

19.07.04

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98165-CLO

## Genetic Discrimination of Prince William Sound Herring Populations

Project Number: 98165 -C L O

Restoration Category: General Restoration

Proposer: ADF&G

Lead Trustee Agency: ADF&G

Cooperating Agencies: University of Washington, Dalhousie University

Duration: 4th year, 4-year project

Cost FY 98: \$56,000

Geographic Area: Prince William Sound

Injured Resource/Service: Pacific herring

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### ABSTRACT

Following the 1989 *Exxon Valdez* oil spill (EVOS), the Prince William Sound herring fishery underwent a catastrophic decline beginning in 1992. Alaska Department of Fish and Game recovery effort includes incorporating a knowledge of genetically derived population structure into harvest management. In this closeout project we delineate the structure of Prince William Sound population(s) and related North Pacific populations using both nuclear and mitochondrial DNA analyses. Results of year-one DNA analyses indicate very limited genetic exchange between the Bering Sea/ Kodiak Island populations and the PWS populations, and there is evidence of significant levels of genetic divergence within PWS.

## INTRODUCTION

Pacific herring *Clupea pallasii* are a major resource in Prince William Sound from both a commercial and ecological perspective. The timing of the *Exxon Valdez* oil spill (EVOS) overlapped the annual spring migration of herring spawners to nearshore staging areas. Over 40% of the herring spawning, staging, and egg deposition areas and over 90% of the documented rearing and feeding areas were lightly to heavily oiled prior to the spawning events. As a result, herring encountered oil during each of their four life stages in 1989 and, to a lesser extent, in 1990. Adult herring traversed oil sheens and mousse while traveling northward and eastward. Eggs were deposited on oiled shorelines and were exposed to sheen through tidal action while incubating. Larvae that hatched contained lipophilic petroleum hydrocarbons in their yolk sacs and encountered sheen near the surface while in their most sensitive state. Post-larval or juvenile herring swam through and remained near lightly to heavily oiled shorelines, regularly encountering sheen, mousse and dissolved oil components through the summer while feeding in shallow nearshore bays and passes.

The Prince William Sound herring fishery underwent a catastrophic decline beginning in 1992. In 1993, the total observed spawning population was less than one-third of preseason predictions, and the average sizes of herring in each age class were some of the smallest on record. Only limited commercial herring fishing occurred. Preliminary pathology results implicated viral hemorrhagic septicemia (VHS) as a potential source of mortality and stress, however this has not been shown conclusively (Meyers et al. 1994). In 1994, as in 1993, the spawning population was below preseason predictions. No recovery was evident in 1995, and based on this, the 1996 commercial fishing season was cancelled. Aerial surveys during 1996 indicated that the population was beginning to recover. In 1997, a commercial herring fishery will occur if aerial surveys indicate the projected biomass of spawning herring materializes. The ex-vessel value of the herring fisheries in 1992 was \$12.0 million. In 1993, the ex-vessel value dropped to \$2.0 million, and herring abundance has been so low that no commercial harvest has occurred since.

Alaska Department of Fish and Game is mobilizing a recovery effort that includes pathology, genetics, early life history, and oceanographic investigations. The Department drafted a stock model (Brown and Wilcock 1994) to provide a basis for restoration management. However, that model is based upon several assumptions about the population structure of and recruitment to Prince William Sound spawning groups. This proposal was designed to evaluate those assumptions which include genetic homogeneity of herring stocks within the Sound and no recruitment to those stocks from outside of the Sound.

Incorporating genetically derived population structure is crucial to the success of any fisheries or restoration program. Consistent exploitation of mixed populations has to lead to the demise of the least productive stocks (Schweigert 1993). Unfortunately, defining the population structure of herring has been particularly difficult. There is evidence that herring home (Wheeler and Winters 1984), but straying may also be substantial. Morphological and meristic differentiation of herring from discrete geographic regions has been used as evidence for the existence of



genetically distinct populations, but much of this variation may be environmentally mediated and has not been confirmed with genetic data (Safford and Boone 1992; King 1985).

Allozyme electrophoresis has proven to be the most useful tool for delineating the population structure of many commercially important species in Alaska. However, previous surveys of herring using this technique have generally revealed differentiation only over broad geographic regions (Grant 1984; Grant and Utter 1984) or between spawning populations within the same area that are temporally isolated (Kornfield et al. 1982). Allozymes defined two distinct races of Pacific herring (Asian/Bering Sea and eastern North Pacific), with further subdivision between the Gulf of Alaska and more southerly North Pacific stocks (Grant and Utter 1984). Also, allozyme markers were used to describe genetic divergence among local spawning populations of Pacific herring in the vicinity of northern Japan (Kobayashi et al. 1990) and among genetically distinct fjord populations in Norway (Jorstad et al. 1994).

Additional techniques to study the structure of natural populations became available in recent years as a result of advances in molecular biology. Restriction fragment length polymorphism (RFLP) analysis of mitochondrial DNA provided some evidence of genetic differentiation among Atlantic and Pacific herring (Kornfield and Bogdanowicz 1987; Schweigert and Withler 1990; Dahle and Eriksen 1990); however the utility of these and more recently developed techniques to detect fine genetic structure in Pacific herring has not been properly assessed. Peer reviewers of preproposal 95165 recommended that, of the molecular techniques then considered by our laboratory, we focus upon microsatellite markers as being potentially the most useful markers for investigation of fine structure. Microsatellites are nuclear loci amplified by PCR which are comprised of regions with variable number of tandem repeats (VNTR) of short base sequences, usually <100bp in total length.

Incorporating peer review comments, and in consideration of the fact that nuclear and mitochondrial loci evolve in response to different pressures, we are pursuing a combination of both mitochondrial and microsatellite approaches to more accurately define the stock structure of herring from the EVOS-affected area (e.g., Taylor and Bentzen 1993; Bentzen et al. 1994). The data may also be used to estimate the population composition of non-spawning aggregations contributing to the fisheries in Prince William Sound. We collaborated under contract (FY95 funds) with the University of Washington Marine Molecular Biotechnology Laboratory and Dalhousie University Marine Gene Probe Laboratory to develop both mtDNA and microsatellite markers for use in examining Pacific herring population structure. These laboratories have developed mtDNA and microsatellite markers and are in the process of finalizing analysis of genetics population data collected from the 1995 samples. Preliminary results appeared promising, and we exercised our option to renew these contracts with FY96 funds for analyses of samples collected during 1996. Analysis of these samples are underway. Thus far in FY97 we have completed sub-sampling and archiving of 1996 herring tissue collections; we contributed a poster in the January 1997 EVOS workshop; we are co-authoring a manuscript of year-one results with our contractors; we are continuing project monitoring; and we will initiate technology transfer to ADF&G late this spring including laboratory analysis of up to 200 fish.

Preliminary results of year-one nuclear DNA analyses (microsatellites) indicate very high levels of genetic diversity. The divergence estimates suggest very limited genetic exchange between the Bering Sea/ Kodiak Island populations and the PWS populations, and there is evidence of significant levels of genetic divergence within PWS (Figure 1). The preliminary mtDNA results also suggest dramatic differentiation between Gulf of Alaska and Bering Sea populations, a high degree of genetic variability in the ND1 gene of Pacific herring, and heterogeneity between Kodiak and PWS populations (Figure 2). Analysis of repeat samples is required to determine if the observed pattern of differentiation is consistent over time.

## **NEED FOR THE PROJECT**

### **A. Statement of Problem**

The Prince William Sound herring fishery is just beginning to recover from a serious decline. The lack of commercial harvest since 1993 has had severe negative impacts on individual fishermen as well as the economies of the communities within Prince William Sound.

### **B. Rationale/Link to Restoration**

Pacific herring are a major resource in Prince William Sound (PWS) from both commercial and ecological perspectives. During the last 15 years the five commercial herring fisheries in PWS had an average annual combined ex-vessel value of \$8.3 million (Donaldson et al. 1993). Pacific herring provide important forage for many species including some species severely injured by the *Exxon Valdez* oil spill. Predator species include humpbacked whales, seals, sea lions, gulls, sea ducks, shorebirds, halibut, salmon, rockfishes, pollock, and other fishes. In addition, several thousand pounds of herring and herring spawn-on-kelp are harvested annually for subsistence purposes and form an important part of the local native culture of the villages of Chenega and Tatitlek.

The goal of this project is to improve the accuracy of current stock assessment methods, thus improving resource management. Improved accuracy of stock distribution information will allow fishery managers to make fine adjustments of fishing quotas to harvest the maximum available surpluses with the lowest possible risk of overharvest, damage to the resource, or economic loss to the fishing industry. This information is also needed to help interpret oil spill damage results.

### **C. Location**

Field research will be conducted primarily within the confines of Prince William Sound; exact locations will depend upon the distribution of spawning herring. Sampling outside of Prince William Sound will be conducted by ADF&G area staff as appropriate. Laboratory sampling, tissue archiving, and data analysis will be conducted at the ADF&G area office in Cordova and

regional office in Anchorage. Prior to the 1998 herring fishing season, meetings will be held with area ADF&G staff in Cordova and Kodiak if changes to fishery management strategies are warranted based on genetics data.

Because commercial and subsistence herring harvests represent substantial contributions to local economies, intensive management is expected to benefit all communities in PWS. Restoration efforts can be directed and evaluated through improved fishery management and continued resource monitoring.

## **COMMUNITY INVOLVEMENT**

Laboratory analyses and reporting are technical pursuits that will be conducted by or supervised by Ph.D. scientists. Wherever possible, local-hire will be used to fill field positions required for sampling or routine laboratory positions.

## **PROJECT DESIGN**

### **A. Objectives**

Our overall objective is to provide a genetic basis for the stock model used by Alaska Department of Fish and Game to manage and restore the severely impacted herring resource in Prince William Sound. We propose to test for genetic heterogeneity among spawning aggregations of Pacific herring within Prince William Sound, adjacent to Prince William Sound, and between year classes within and adjacent to the Sound. Achieving this objective will provide information to enable resource managers to better understand herring population dynamics and make management decisions to speed the recovery process. In addition, it will aid local resource users to make appropriate pre-season plans based on accurate and precise herring projections.

The working objectives of this study are to:

1. Screen population samples using both mitochondrial and nuclear DNA approaches. Techniques will include RFLP analysis of mitochondrial DNA and microsatellite analysis of nuclear loci.
2. Evaluate the null hypothesis that a single panmictic population of herring exists in Prince William Sound. The study will include at least four putative population samples from both spatial and temporal isolates within the Sound.
3. Evaluate the structure of Prince William Sound herring populations within the context of the structure of adjacent spawning aggregates (up to four), including comparisons from across the known genetic barrier of the Alaska Peninsula.

4. Test for inter-annual stability of allele frequencies in Prince William Sound and related North Pacific populations.

## B. Methods

### 1. Field Collections

Earlier versions of this proposed project focused solely upon populations within Prince William Sound. Peer reviewers recommended expanding the project to include outside stocks for comparison from the Gulf of Alaska and the Bering Sea and to include tests for inter-annual stability (cf., Kornfield et al. 1982, see below).

During 1995, field collections of spawning Pacific herring targeted eight representative sites within and adjacent to Prince William Sound (Table 1, Figure 3). The collection sites within Prince William Sound were chosen to maximize the potential genetic differentiation among temporally and spatially isolated spawning aggregations. Tissue extracts from muscle, liver, eye, and heart were collected and preserved in liquid nitrogen for transport to  $-80^{\circ}\text{C}$  freezers for archiving. A second year of sampling (1996) was conducted at each site to test for inter-annual stability of gene frequencies (Table 1, Figure 3).

The within-Sound sampling efforts in 1995 targeted Rocky Bay, a southcentral spawning isolate on Montague Island; St. Matthews Bay, a southeast isolate; Fish Bay, a northeast isolate, and Port Chalmers on Montague Island (Figure 3). Efforts to sample both early- and late-spawning stocks within these four sites were unsuccessful in 1995 due to the timing of the spawning returns and inclement weather conditions which hindered early collection efforts. Single collections were made in these spawning sites. Early-spawning isolates were collected from St. Matthews Bay and Fish Bay, and late-spawning isolates were collected from Rocky Bay and Port Chalmers. Collection plans for 1996 included resampling of St. Matthews Bay, Fish Bay, Rocky Bay and Port Chalmers for both early- and late-spawning stocks, and a collection from Kayak Island in PWS. However, the timing of spawning returns to PWS in 1996 precluded collection of temporal samples. Single collections were made in St. Matthews Bay, Fish Bay, Rocky Bay, and Stockdale Harbor near Pt. Chalmers (Table 1).

One-hundred individuals were subsampled from each aggregation during the sampling for Trustee Council Project 97166 *Herring Natal Habitat*. Consequently, age and other data will become available for many of the individuals analyzed for genetic variation, facilitating further correlation analyses between population data and genetic variation.

Sampling outside of Prince William Sound in 1996 included Kodiak Island, populations thought to share an ancestral tie with Prince William Sound populations (John Wilcock, Alaska Department of Game, personal communication) and Bering Sea populations

known to be genetically isolated from the other Gulf of Alaska stocks (Grant and Utter 1984; Figure 3). During 1996, we resampled the three outside stocks included for comparison in the FY95 initial laboratory analyses (Kodiak, Togiak, Norton Sound). Additional outside stock sample collections were made from Sitka Sound and Port Moller. Muscle tissue from 100 individuals were collected from each of these two populations.

## 2. Genetic Analysis

The preproposal for this project included allozyme analysis as well as DNA analysis, however peer reviewers recommended that year-one of the study focus on techniques such as microsatellite analysis to maximize the probability of identifying genetic differences (as described herein). Through further public review we decided to collect and archive samples in 1995 for allozyme analysis because the area affected by EVOS is adjacent to the genetic barrier zone identified by allozymes and the loss of the opportunity to compare allozyme results to DNA results would be irretrievable (W. S. Grant, National Marine Fisheries Service, personal communication). Allozyme-quality tissues collected in 1995 were archived at -80° C.

Alaska Department of Fish and Game solicited assistance from outside laboratories for the genetic analyses following standard State of Alaska procurement procedures for Project 95165 analyses. A request for proposal was issued for the molecular analyses, and contracts granted to two university laboratories. At this writing ADF&G is working under contract with Dr. Paul Bentzen at the University of Washington for mtDNA and Dr. Jonathan Wright at Dalhousie University for microsatellite marker analyses under Trustee Council approved Project 96165 funding. Because of the timing of the awarding of this contract, final results are pending, and analysis of 1996 samples is underway. However, preliminary results were provided in the 95165 annual report to the Trustee Council, and in the 1996 and 1997 EVOS January workshops.

We chose the current contract laboratories for their joint proposal which incorporated both mtDNA and microsatellite analyses. The Principal Investigators in these laboratories have published extensively in this area, applying both mtDNA and microsatellite methods to questions of population structure (e.g. Roff and Bentzen 1989; Bentzen et al. 1991; Bentzen et al. 1993a; Bentzen et al. 1993b; Bentzen and Wright 1993; Wright 1993; Bentzen et al. 1994; Wright and Bentzen 1994; Morris and Wright 1996).

Alaska Department of Fish and Game evaluated the preliminary results of project 95165 and opted to renew the current contracts for one additional year of sample analysis under project 96165. Depending on the final results of the 95165 analyses and preliminary results of 96165 sample analyses, we will likely exercise the option of conducting laboratory analysis of a small number (~200) of additional PWS samples and/or outside

stock comparative samples during the technology transfer to ADF&G..

### **C. Cooperating Agencies, Contracts and Other Agency Assistance**

The Alaska Department of Fish & Game will initiate transfer of herring microsatellite technology developed by Dalhousie University while still under contract (96165) with a one week on-site consultation with Dr. Jonathan Wright. Transfer of herring mtDNA technology developed by the University of Washington to ADF&G will also be initiated by a one week on-site consultation with Dr. Paul Bentzen if needed.

Contracting laboratory analysis will become less efficient for the Department as other projects currently underway at ADF&G reach completion. Future contracts to other laboratories will only be awarded if the work cannot be done in-house.

## **SCHEDULE**

### **A. Measurable Project Tasks for FY 97 and FY98**

October 1, 1996:	Tissue sub-sampling and archiving; begin lab analyses of FY96 samples
January - June, 1997:	Evaluate final FY95 lab results, initiate technology transfer
April 15, 1997:	Proposal for FY98
March - June 15, 1997:	Collection of mop-up samples if needed
June 1- Sept. 30, 1997:	Technology transfer; conduct mop-up sampling and sample re-runs
September 30, 1997:	close out FY96 funded contracts
October 1, 1997-	
September 30, 1998:	Conclude technology transfer; conclude laboratory analysis of remaining FY96 and FY97 samples; data analyses
September 30, 1998:	Data analyses and final report

### **B. Project Milestones and Endpoints**

September 30, 1997:	Complete survey of population samples collected during 1996 and 1997; evaluate stability of population structure across years; complete technology transfer
October 1, 1997 -	
March 30, 1998:	Conclude in-house sample analyses
September 30, 1998:	Close out Project 98165

### **C. Completion Date**

Project 165 is anticipated to be completed by September 30, 1998.

### **PUBLICATIONS AND REPORTS**

September 30, 1997: contractors reports FY96

June 1997: year-one microsatellite data report in the form of manuscript submitted to the Canadian Journal of Fisheries and Aquatic Sciences

September 30, 1998: final report in the form of manuscript submitted to journal

### **PROFESSIONAL CONFERENCES**

AFS-Alaska Chapter; Sitka AK., November , 1997; Genetic structure of herring populations

### **NORMAL AGENCY MANAGEMENT**

The Alaska Department of Fish and Game spends approximately \$500.0K from State of Alaska general funds annually on genetics studies. For this project, salaries and benefits of principal investigators J. Seeb and L. Seeb are fully funded by general funds; project leader S. Merkouris is funded for three months from Trustee Council funds and three months by general funds. These general funds from the legislature are ear-marked for specific projects; although they may be used for leadership of EVOS studies, no general funds are available to institute new research such as this. The *Exxon Valdez* Trustee Council has shouldered the burden of research into the ecology and genetics of species within the spill zone. These studies would not have been conducted by the State in the absence of the oil spill.

The Department remains heavily committed to the conduct of this study and other EVOS studies, even though limited personnel resources mandate that we seek assistance from an outside source for the FY95 and FY96 laboratory analyses described herein. State of Alaska general funds support the basic operation of and enhancements to the genetics laboratory for EVOS projects including the procurement of an Applied Biosystems Incorporated automated DNA sequencing system capable of subambient temperature operation required for studies of genetic variation including RFLP analysis (\$132.0K).

Staff scientists and technicians are trained in an array of genetics analyses including allozyme and PCR-based mitochondrial and nuclear approaches. The Department maintains fourteen -80° C freezers in area offices throughout the state for archival of genetic samples for allozyme and DNA analyses.

## **COORDINATION AND INTEGRATION OF RESTORATION EFFORT**

Collection of specimens and biological data will be coordinated by ADF&G's ongoing herring research program in Prince William Sound and with the EVOS project 98166 Herring Natal Histories. Tissue archival and biometric analyses will be coordinated among all Trustee Council projects related to genetics including 98196 and 98252.

Sharing of project results will be used to evaluate and revise current strategies for management of commercial herring fisheries if warranted. Dissemination of these data will occur during on-sites meeting with area fishery managers and researchers in Cordova and Kodiak. Project results will also be used to improve our understanding of results from previous oil spill damage assessment studies.

Data collection techniques will be coordinated through the inter-agency consortium of laboratories that cooperate on similar projects of conservation genetics of marine fishes in the North Pacific Ocean.

## **EXPLANATION OF CHANGES IN CONTINUING PROJECTS**

The duration of this project was originally anticipated to be two and one-half years (see Projects 94165 and 95165). This period was to cover field collections from two spawning seasons and subsequent laboratory analysis. We anticipated that laboratory analysis would be complete in FY95 and reporting to be complete in FY96.

However the start date, and thus the completion date, of this project have been elusive. The Trustee Council first made funds available during FY94 (Project 94165). The field season that year was truncated due to the surprise run failure, inadequate samples were obtained to meet most project objectives, and project start was deferred one year. No Trustee Council funds were spent on the project in FY94. Contract lab analyses of FY96 samples (Project 96165) are underway at the writing of this proposal, 98165. Spatial isolates from within the Sound were successfully sampled during 1995 and 1996, but temporal isolates were elusive because of the run failure. Sampling from outside of the Sound was successful in 1995 and 1996. We initiated laboratory analyses with the aid of a contractor in 1995, but FY95 sample collections alone were not adequate to meet all four project objectives.

At least two years of complete sampling and analyses are required to confirm interannual stability of population structure. For example, Kornfield et al. (1982) observed ~~within-year~~ temporal variation and within-year spatial variation in Atlantic herring populations that were not stable across year classes. Such annual variation may indicate substructure variability due to changes in larval flushing/larval retention patterns such as those described in Brown and Wilcock (1994). Thus, management recommendations made on only one year's genetic data may not be



valid. Based upon sampling difficulties due to the run failure, we now believe that reporting of this project will not be complete until FY 98. The cover sheet for this proposal reflects a FY98 budget request for data analyses and reporting close-out budget for FY 98 is also included.

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## PROPOSED PRINCIPAL INVESTIGATORS

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PROJECT RESPONSIBILITIES: Design, analysis, reporting

### EDUCATION:

B.S., Biology, 1974, University of Puget Sound

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### PROFESSIONAL EXPERIENCE:

1990- Principal Geneticist, CFMD Division, ADF&G  
1988-1990 Assistant Professor, Southern Illinois University  
1987-1988 Research Assistant Professor, University of Idaho  
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**B. Lisa W. Seeb (L. Wishard), Statewide Geneticist**  
Division of Commercial Fisheries Management and Development  
Alaska Dept. of Fish and Game  
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PROJECT RESPONSIBILITIES: Biometrics, analysis, reporting

EDUCATION:

Prepared April 15, 1996

16

Project 98165

A.B. Zoology, 1973, University of California, Berkeley

M.A. Zoology, 1977, University of Montana

Ph.D. Fisheries, 1986, University of Washington

#### PROFESSIONAL EXPERIENCE:

1991-           Statewide Geneticist, ADF&G, Anchorage  
1991-           Affiliate Associate Professor, U. of Alaska, Fairbanks  
1988-1990     Assistant Professor, Southern Illinois University  
1984-1988     Research Assist. Prof., University of Idaho  
1978-1981     Fish Geneticist, Pacific Fish. Research, Olympia WA  
1977-1979     Geneticist, National Marine Fisheries Service, Seattle

#### SELECTED PUBLICATIONS:

Wishard, L. N., J. E. Seeb, F. M. Utter, and D. Stefan. 1984. A genetic investigation of suspected redband trout populations. *Copeia* 1984(1):120-132.

Seeb, J. E., L. W. Seeb, and F. M. Utter, 1986. Use of genetic marks to assess stock dynamics and management programs for chum salmon. *Trans. Amer. Fish. Soc.* 115:448-454

Seeb, L. W. and D. R. Gunderson. 1988. Genetic variation and population structure of Pacific ocean perch (*Sebastes alutus*). *Can. J. Fish. Aquat. Sci.* 45:78-88.

Seeb, L. W., J. E. Seeb, R. L. Allen and W. K. Hershberger. 1990. Evaluation of adult returns of genetically marked chum salmon, with suggested future applications. *American Fisheries Society Symposium* 7:418-425

Seeb, L. W., J. E. Seeb and A. J. Gharrett. 1990. Genetic marking of fish populations. pp 223-239 in D. H. Whitmore, ed. *Electrophoretic and isoelectric focusing techniques in fisheries management*. CRC Press, Boca Raton, FL.

Seeb, L. W., J. E. Seeb and J. J. Polovina. 1990. Genetic variation in highly exploited spiny lobster *Panulirus marginatus* populations from the Hawaiian Archipelago. *Fishery Bulletin* 88:713-718.

Seeb, L. W. and A. W. Kendall. 1991. Allozyme polymorphisms permit the identification of larval and juvenile rockfishes of the genus *Sebastes*. *Environmental Biology of Fishes* 30:191-201.

Utter, F. M., J. E. Seeb, and L. W. Seeb. 1993. Complementary uses of ecological and biochemical genetic data in identifying and conserving salmon populations. *Fisheries Research*. *Fish. Res.* 18:59-76.

Crane, P. A., L. W. Seeb, and J. E. Seeb. 1994. Genetic relationships among *Salvelinus* species inferred from allozyme data. Can. J. Fish. Aquat. Sci. 51(Suppl. 1):182-197.

Seeb, L. W. C. Habicht W. D. Templin, K. E. Tarbox R. Z. Davis, L. K. Brannian, J. E. Seeb. 1998. Genetic diversity of sockeye salmon (*Oncorhynchus nerka*) of Cook Inlet, Alaska, and its application to restoration of populations affected by the Exxon Valdez oil spill. Canadian Journal of Fisheries and Aquatic Sciences. *Accepted and in press.*

Seeb, J. E. C. Habicht, J. B. Olsen, and L. W. Seeb. 1998. An overview of gene detection methods used to study population variation in salmonids. Assessment and Status of Pacific Rim Salmonid Stocks. North Pacific Fish Commission, Vancouver B.C. *Accepted and in press.*

**C. Susan E. Merkouris**, Shellfish and Marine Fishes Project Geneticist  
Commercial Fisheries Management and Development  
Alaska Department of Fish and Game  
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**PROJECT RESPONSIBILITIES:** Field coordination, sampling, archiving, contracting, laboratory and data analyses, reporting

**EDUCATION:**

A.A., 1974, Liberal Arts (Honors), Golden Valley Lutheran College, Mpls., MN  
B.S., 1980, Biology and Chemistry, *magna cum laude*, University of Alaska, Anchorage AK

**PROFESSIONAL EXPERIENCE:**

1991- Shellfish and Marine Fishes Project Geneticist, CFMD, ADF&G  
1989-1991 Lower Yukon Asst. Mgmt. Fisheries Biologist, C.F., ADF&G  
1985-1989 Norton Sound Asst. Mgmt. Fisheries Biologist, C.F., ADF&G  
1981-1985 Fisheries Biologist, C.F., ADF&G  
1979-1981 Fisheries Technician, C.F., ADF&G  
1976-1980 Clinical Microbiologist, Norton Sd.Regional Hospital, Nome, AK

**SELECTED PUBLICATIONS AND PRESENTATIONS:**

Merkouris, S. E. and L. W. Seeb. (accepted pending revisions). Genetic variation of highly exploited Tanner crabs, *Chionoecetes bairdi* and snow crabs, *C. opilio* in Alaska. Submitted to Fishery Bulletin.



Seeb, L. W. and S. E. Merkouris. (in prep). Hybridization between highly exploited tanner and snow crabs, *Chionoecetes bairdi* and *C. opilio*, in the Bering Sea. Preliminary results presented at Genetics of Subarctic Fish and Shellfish International Symposium, Juneau, AK, 1993.

Merkouris, S. E. and C. F. Lean. 1989. Annual Management Report 1988 Norton Sound - Port Clarence - Kotzebue. Alaska Department of Fish and Game, Regional Information Report Series. RIR #3A89-10.

Bergstrom, D. J., S. E. Merkouris, K. Schlutz, R. Holder, G. Sandone, D. Schneiderhan, L. H. Barton, and D. Mesiar. 1991. Annual Management Report Yukon Area, 1989. Alaska Department of Fish and Game, Regional Information Report Series. RIR #3A91-14.

Table 1. Description of Pacific herring samples collected for Project 165. Location number corresponds to Figure 3.

Location Number	N	Dates Sampled	Lat. N.	Long. W.	Location and timing <sup>1</sup>
1	100	4/95	60°42'	146°20'	St. Matthews Bay, early
	100	4/96	60°42'	146°20'	St. Matthews Bay, early
2	100	4/95	60°49'	146°25'	Fish Bay, early
	100	4/96	60°49'	146°25'	Fish Bay, early
3	100	4/95	60°21'	147°07'	Rocky Bay, late
	100	4/96	60°21'	147°07'	Rocky Bay, late
4	100	4/95	60°15'	147°13'	Port Chalmers, late
	100	4/96	60°19'	147°13'	Port Chalmers, late <sup>2</sup>
5	100	3/96	57°00'	135°30'	Sitka Sound
6	90	5/95	58°06'	153°04'	Kodiak Island
	100	5/96	57°46'	153°12'	Kodiak Island <sup>3</sup>
7	100	5/96	56°04'	160°45'	Port Moller
8	100	5/91	58°50'	160°24'	Togiak Bay
	100	5/96	58°50'	160°24'	Togiak Bay
9	100	5/91	63°54'	160°50'	Norton Sound
	100	5/96	63°54'	160°50'	Norton Sound

<sup>1</sup> The absence of bimodal spawning activity precluded within-year temporal sampling. 'Early' and 'late' refer to timing of spawning relative to all of PWS within a collection year.

<sup>2</sup> Fish collected in Stockdale Harbor are believed to have spawned in Port Chalmers.

<sup>3</sup> Fish did not spawn in Muskomee Bay (Raspberry Strait) in 1996, the 1995 collection site. The 1996 samples were collected from spawning fish in Terror Bay, Uganik Passage, also on westside Kodiak Island.

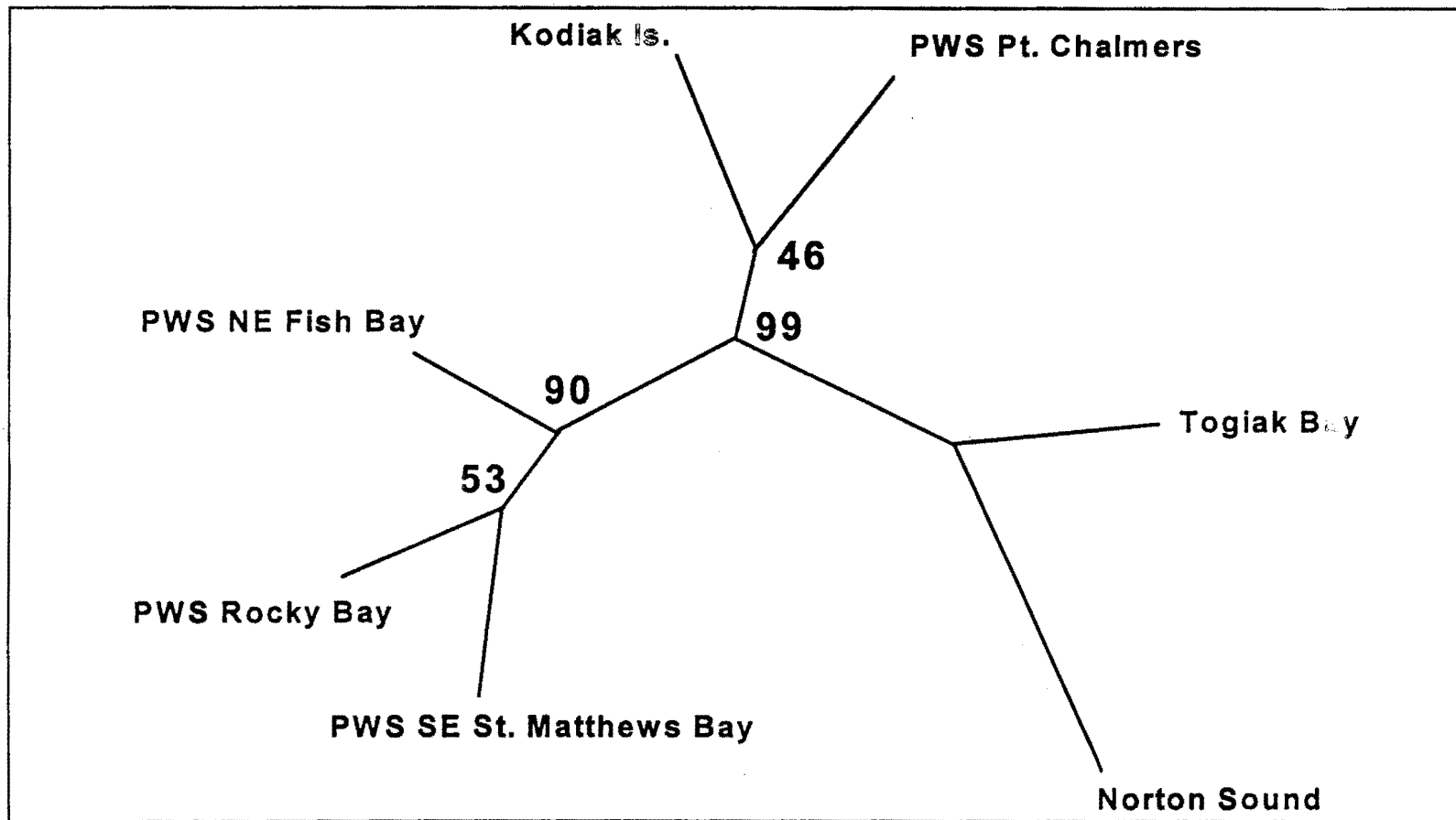


Figure 1. Maximum likelihood tree constructed from allele frequencies of five microsatellite loci (Felsenstein 1989). Bootstrap values were calculated over 1,000 replications by re-sampling across loci.

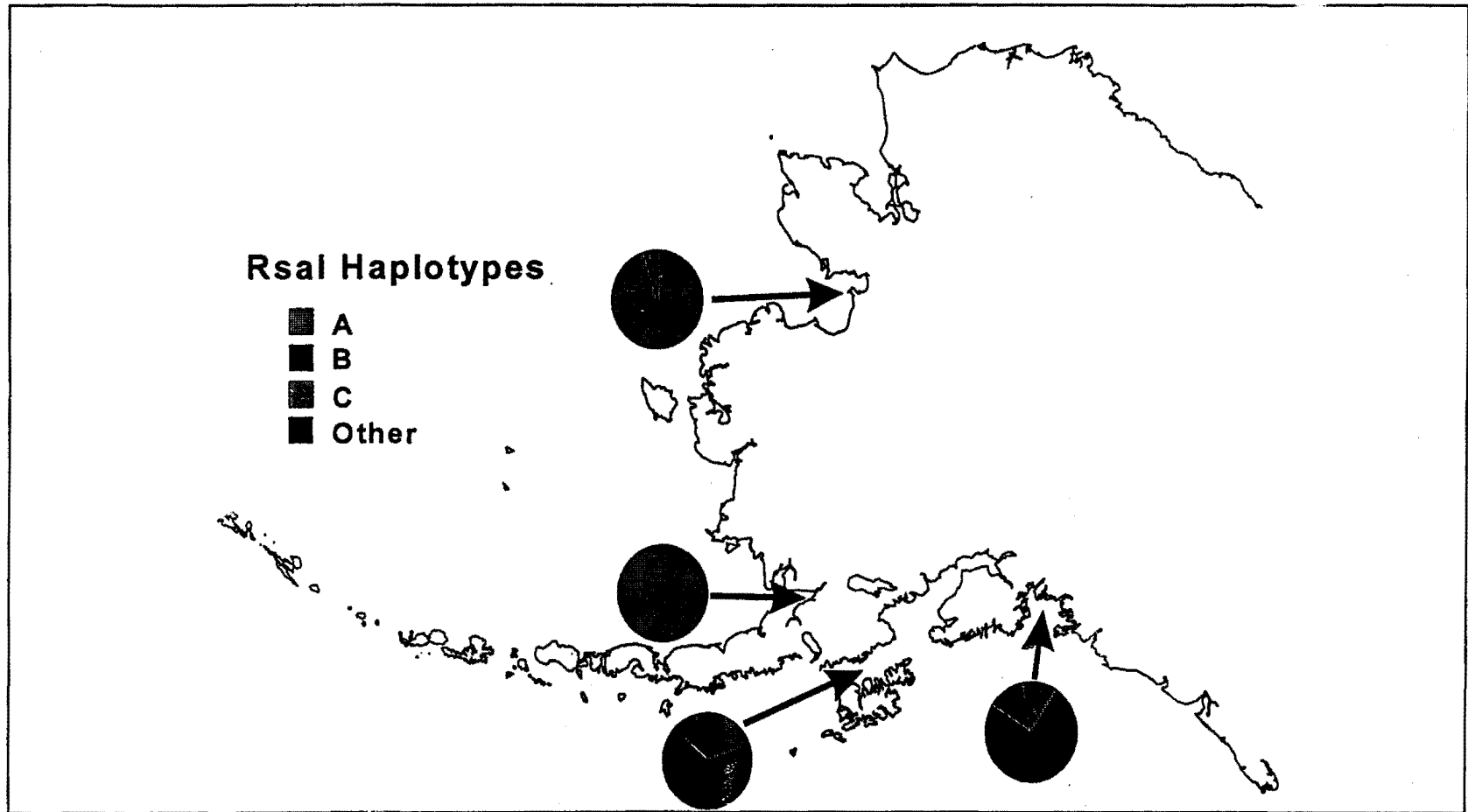


Figure 2. *RsaI* haplotype frequencies of mtDNA-ND1 in Gulf of Alaska and Bering Sea herring populations. The pie for Prince

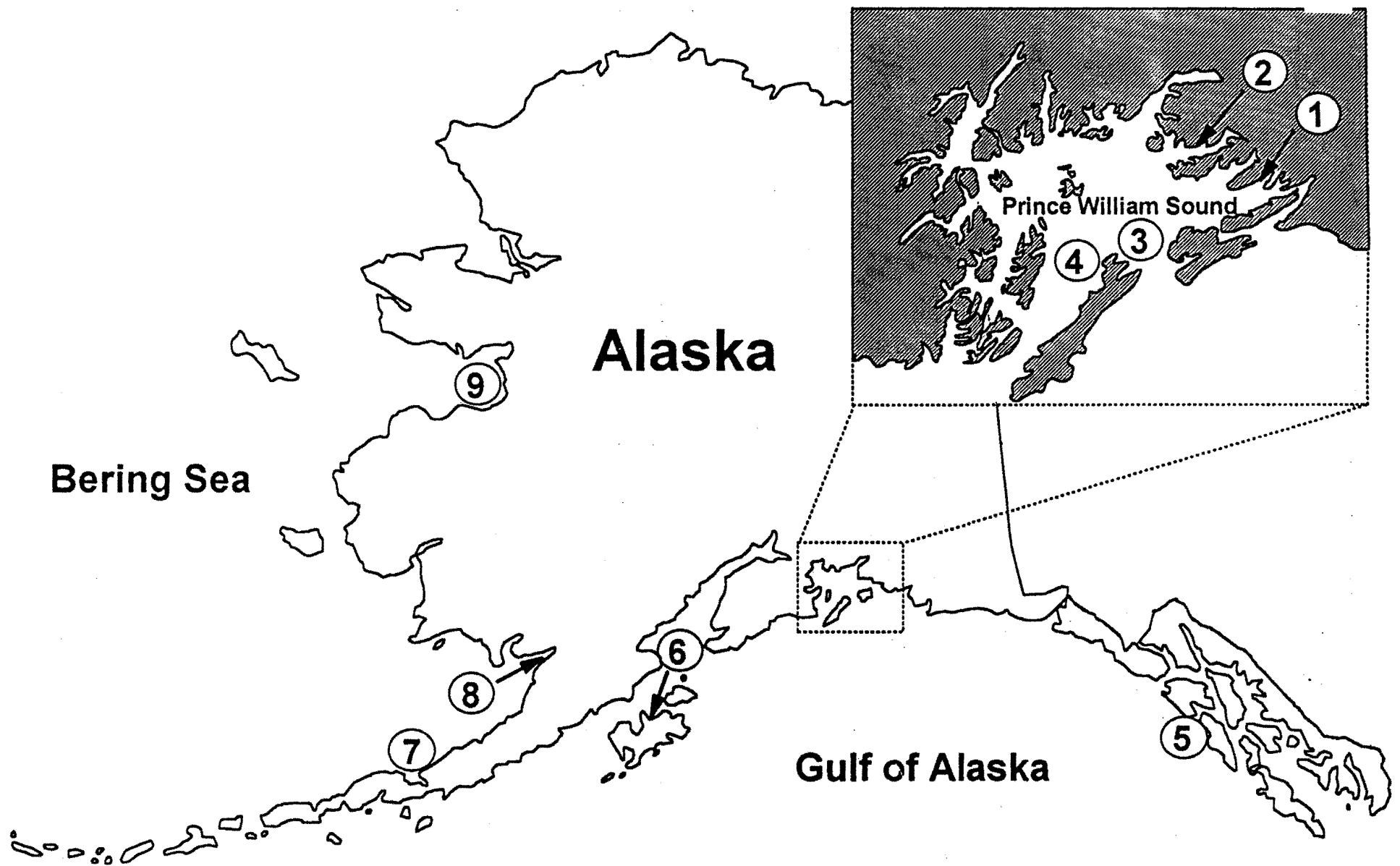


Figure 3. Prince William Sound, Gulf of Alaska, and Bering Sea sample collection sites for 1995 and 1996

# 1998 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1997 - September 30, 1998

Budget Category:	Authorized FFY 1997	Proposed FFY 1998						
Personnel	\$23.0	\$28.5						
Travel	\$6.0	\$5.2						
Contractual	\$2.8	\$6.9						
Commodities	\$6.2	\$10.6						
Equipment	\$0.0	\$0.0	LONG RANGE FUNDING REQUIREMENTS					
Subtotal	\$38.0	\$51.2	Estimated FFY 1999	Estimated FFY 2000	Estimated FFY 2001	Estimated FFY 2002	Estimated FFY 2003	
General Administration	\$3.6	\$4.8						
Project Total	\$41.6	\$56.0						
Full-time Equivalents (FTE)		0.5						
Dollar amounts are shown in thousands of dollars.								
Other Resources								
Comments:								

1998

Project Number: 98165 - CLO  
 Project Title: Genetic discrimination of PWS herring populations  
 Agency: AK Dept. of Fish & Game

FORM 3A  
 TRUSTEE  
 AGENCY  
 SUMMARY

**1998 EXXON VALDEZ TRUS. ... COUNCIL PROJECT BUDGET**

October 1, 1997 - September 30, 1998

Personnel Costs:		GS/Range/ Step	Months Budgeted	Monthly Costs	Overtime	Proposed FFY 1997
Name	Position Description					
Merkouris	FBI (PCN 1390)	16K	3.0	5.5		16.5
Kretschmer	FBI (PCN 7112)	14B	3.0	4.0		12.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
Subtotal			6.0	9.5	0.0	
Personnel Total						\$28.5
Travel Costs:		Ticket Price	Round Trips	Total Days	Daily Per Diem	Proposed FFY 1997
Description						
Anch to Cordova, full fare		0.22	3			0.7
Anch to Kodiak, full fare		0.22	3			0.7
Per diem				10	0.15	1.5
						0.0
Anch to Sitka, Professional Meeting		0.50	1			0.5
Per diem				4	0.15	0.6
						0.0
Anch to Seattle, Professional Meeting		0.40	1			0.4
Per diem				5	0.15	0.8
						0.0
						0.0
						0.0
Travel Total						\$5.2

**1998**

Project Number: 98165 - CLO  
 Project Title: Genetic discrimination of PWS herring populations  
 Agency: AK Dept. of Fish & Game

FORM 3B  
 Personnel  
 & Travel  
 DETAIL

Prepared: 2 of 4

4/15/97

# 1998 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1997 - September 30, 1998

Contractual Costs:		Proposed
Description		FFY 1998
Aircraft charter		0.0
Air freight/DHL		0.5
Photographic development		0.5
DNA Sequencer maintenance contract, equipment repair		4.5
Reproduction / printing costs		0.7
Telephone charges		0.7
When a non-trustee organization is used, the form 4A is required.		
Contractual Total		\$6.9
Commodities Costs:		Proposed
Description		FFY 1998
Sampling, archiving supplies		0.6
Laboratory supplies		2.0
Biochemicals, DNA enzymes, primers		7.5
Office / presentation supplies		0.5
Commodities Total		\$10.6

1998

Project Number: 98165 - CLO  
 Project Title: Genetic discrimination of PWS herring populations  
 Agency: AK Dept. of Fish & Game

FORM 3B  
 Contractual  
 & Commodi  
 ties



**1998 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET**  
October 1, 1997 - September 30, 1998

[illegible]

1998

Project Number: 98165 - CLO  
Project Title: Genetic discrimination of PWS herring populations  
Agency: AK Dept. of Fish & Game

FORM 3B  
Equipment  
DETAIL



**EXXON VALDEZ Oil Spill Trustee Council**  
**FY 98 Detailed Project Description**

**Herring Natal Habitats**

<b>Project Number:</b>	98166
<b>Restoration Category:</b>	General Restoration
<b>Proposer:</b>	ADF&G
<b>Lead Trustee Agency:</b>	ADF&G
<b>Cooperating Agencies:</b>	University of Alaska
<b>Alaska SeaLife Center:</b>	
<b>Duration:</b>	Close-out in FY99. Transition to normal agency funding after FY98.
<b>Cost FY 98:</b>	\$189,700
<b>Cost FY 99:</b>	\$22,400
<b>Cost FY 00:</b>	\$0
<b>Geographic area:</b>	Prince William Sound
<b>Injured Resource/Service:</b>	Pacific Herring

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**EXXON VALDEZ OIL SPILL  
TRUSTEE COUNCIL**

**ABSTRACT**

The 1989 *Exxon Valdez* oil spill coincided with the spring migration of Pacific herring *Clupea pallasii* to spawning grounds in Prince William Sound (PWS). Studies of oil spill injuries to herring documented damage from oil exposure in adult herring, reduced hatching success of embryos, and elevated levels of physical and genetic abnormalities in newly hatched larvae. The PWS herring spawning population has drastically declined since 1993, and pathology studies implicated viral hemorrhagic septicemia (VHS) and *ichthyophonus* as potential sources of mortality as well as indicators of stress. The current project will monitor the abundance of the injured herring resource in PWS using spawn deposition techniques. Normal agency funding will be used to conduct acoustic biomass survey. In addition, we will evaluate the precision, accuracy and cost of each technique with the intent to employ either spawn deposition or hydroacoustics using normal agency funding after FY98.

## INTRODUCTION

The *Exxon Valdez* oil spill (EVOS) coincided with the spring migration of Pacific herring *Clupea pallasii* to spawning grounds in Prince William Sound (PWS). Adult herring swam through oiled waters on their way to nearshore staging areas. Studies of oil spill injuries to herring were initiated in 1989 and research continued through 1992. Significant histopathological damage was measured in adults collected in oiled areas in both 1989 and 1990 confirming exposure of the fish to toxins (Brown 1995). Oiling of over 40% of the spawning areas (42 of 98 miles) caused elevated levels of physical and genetic abnormalities in newly hatched larvae and reduced hatching success of the embryos (Brown 1995). Over 80% of the summer rearing and feeding areas of herring were oiled in 1989, based on oil trajectory and historic fisheries records from 1914 to the present (Brown 1995).

In 1993, the herring population in PWS collapsed. The total observed spawning population was less than one third of preseason predictions and the average sizes of herring in each age class were some of the smallest on record. The total commercial harvest for that year was one of the lowest on record. In 1994, the total observed spawning population was below threshold biomass required to conduct a commercial harvest and no fishing occurred. Pathology studies implicated viral hemorrhagic septicemia (Meyers et al. 1994) and a second potentially lethal pathogen, *ichthyophonus*, as possible sources of mortality and stress. Pathology studies continued in 1995 included laboratory investigations of the lethality of suspect pathogens and the role of environmental contaminants in disease transmission.

This project will provide a direct measure of adult herring abundance necessary for monitoring recovery of the injured PWS herring population. ADF&G will perform the field collection and data analysis constituting the continuation of herring spawn deposition surveys. A second field component will investigate the feasibility and cost effectiveness of estimating biomass of spawning herring using acoustic surveys as an alternative to spawn deposition surveys. Acoustic surveys will be subcontracted through a competitive bid process and will rely on ADF&G base funding for much of the vessel and personnel costs.

During spawn deposition surveys, SCUBA divers will estimate the abundance and distribution of herring eggs. This information will be incorporated with aerial observations of spawn distribution and basic biological information (age composition, sex ratios, average size, and fecundity) to estimate adult spawning biomass. Estimates of spawning biomass are used to forecast spawning returns the following year and form the basis of herring fishery management in PWS.

Biomass of herring migrating to PWS spawning grounds will also be estimated acoustically using echo integration techniques. Dual or split beam *in situ* measurements and fish species composition and average size from seine hauls will be used to evaluate and correct for target strength assumptions. Acoustic biomass estimates will be compared with spawn deposition biomass estimates to evaluate the accuracy, reliability, and cost effectiveness of each method.

## NEED FOR PROJECT

### A. Statement of Problem

Adult Pacific herring on their way to PWS spawning areas swam through oil from the *T/V Exxon Valdez* oil spill, eggs incubated in the oil, and larvae and juvenile herring may have been exposed to oil in rearing and feeding areas. Histopathological damage was found in adult herring collected in oiled areas in both 1989 and 1990, mortality of young herring was significantly greater in oiled areas in 1989 and 1990, and sublethal effects were measurable in larvae and adults in 1989 and 1990 (Brown 1995). Persistent sheening and suspended oil-sediment droplets leaching from beaches and cleaning operations in 1989 and 1990 continued to expose adult and juvenile herring to oil. Laboratory exposures of pre-spawning adult herring to oil show high concentrations of oil in the ovarian tissue (Brown 1995). Laboratory studies measuring the effect of known doses of oil on newly hatched larvae provided a direct link between estimated doses of oil measured in PWS and the level of injury observed in samples collected from the field (Brown 1995). In addition, measurements of oil in mussel tissue collected adjacent to spawning beds was significantly correlated to several indices of injury in herring larvae from those beds, the highest correlation being with the genetic injury endpoints (Brown 1995).

Although herring survival varies tremendously under normal conditions, abundance for the 1989 year class was extremely low and results to date strongly implicate the oil spill as a major cause. One hypothesis is that injury to germ tissue caused by exposure to oil may have resulted in non-viable embryos and larvae. A pilot experiment to measure the ability of herring from this age class to produce viable offspring was conducted in 1992 and hatching success of eggs collected from fish spawning in previously oiled areas was less than half that of eggs collected from fish spawning in pristine areas (Brown 1995). Additionally, there were approximately twice as many abnormal larvae from fish spawning in previously oiled areas (Brown 1995).

In 1993, the total observed spawning population was less than one third of preseason predictions and the average sizes of herring in each age class were some of the smallest on record. The total commercial harvest for that year was one of the lowest on record. Pathology studies from the spring of 1993 implicated viral hemorrhagic septicemia (VHS) as a potential source of mortality and stress (Meyers et al. 1994). In 1994 and 1995, the total observed spawning population was below threshold biomass required to conduct commercial harvest and no fishing occurred. Later, pathology studies indicated the presence of both VHS and a second potentially lethal pathogen, *ichthyophonus*. Pathology studies continued in 1997 include laboratory investigations of the lethality of suspect pathogens and the role of environmental contaminants in disease transmission.

## **B. Rationale/Link to Restoration**

The two overall goals of this project have been to monitor recovery of the PWS herring resource which was injured by the EVOS, and to aid in its restoration through improved management of human usage. The Herring Natal Habitats project has accomplished this by improving our ability to more accurately and precisely assess the herring population spawning within PWS (i.e. it provides a population estimate that is within 25% of the true biomass 95% of the time). In addition, normal agency funding will be used for development of hydroacoustic assessment techniques that may provide a similar level of accuracy and precision at a lower cost. Without this project, our only other measure of spawning population biomass is from aerial surveys.

In 1994, the Alaska Board of Fisheries (BOF) established a threshold of 22,000 tons below which a commercial herring harvest would not occur in PWS. This 'productivity' threshold is based upon a simulation of the PWS herring population over a 1000 year period using recruitment, growth, maturity, and natural mortality parameters estimated for this population since the mid 1970's (Zheng et al. 1993). A productivity threshold is defined in terms of quickly rebuilding a population to a commercially productive level (Funk and Rowell 1995). The productivity threshold for PWS herring was set at 25% of the average unfished biomass over the simulation. The simulation indicated that eliminating harvest below the threshold would reduce the risk of population collapse and increase long-term productivity (Zheng et al. 1993). The forecasted PWS herring biomass in 1997 was 34,000 tons.

Zheng et al. (1993) concluded that the success of threshold management strategies is highly dependent on the accuracy of population estimates. Systematic bias may occur from aerial surveys, because peak aerial survey biomass may represent a fraction of total biomass. This potential bias is due to extended migrations to and from the spawning grounds and a reduction of visibility during periods of poor weather. Independent biomass estimates are integral components of the age-structured analysis models used to forecast herring abundance and set harvest levels. A relatively accurate and precise biomass estimate, such as that provided by spawn deposition surveys, is needed to "scale" abundance trends based on age- composition information within the model. Clearly, management precision will be reduced if the greater level of accuracy and precision provided by the spawn deposition biomass estimates is not available. A reduction in management precision when the population is near the productivity threshold may lead to inappropriate harvest levels causing a delay in resource recovery. Management of human use is the most direct action we can take to effect recovery of a depressed resource.

## **C. Location**

This project will be conducted entirely within PWS. Project results will directly affect the management of PWS herring fisheries. All major PWS communities, including Cordova, Seward, Valdez and Whittier, are directly affected by these fisheries since these communities house not only commercial fishers but also the various support services relating to vessel and

gear repair and storage, as well as fish processing. Many native villages in PWS, such as Tatitlek and Chenega, also depend upon PWS herring for subsistence needs.

## COMMUNITY INVOLVEMENT

Since the dramatic decline of the PWS herring spawning population in 1993 there has been vigorous public support for herring research from PWS communities as well as various private and professional organizations. The Public Advisory Group (PAG) for the Trustee Council has also voiced support for these studies. Spawn deposition surveys have been recognized by commercial fishermen, fishery managers, and peer reviewers as a valuable tool for stock assessment in the absence of direct methods of estimation. Accurate and precise estimates of stock abundance are needed for ecosystem based studies of processes that affect abundance. In addition to peer review through the EVOS process, herring stock assessment and embryo survival studies have received critical review through the intensive SEA research planning and public review effort. The ecosystem approach to PWS studies adopted by the SEA planning group recognized the commercial and ecosystem importance of herring and included them as a co-target species for study along with pink salmon.

Some people from communities in PWS will have an opportunity to directly participate in this project by providing logistical support for field sampling. One vessel will be chartered as a research platform for spawn deposition surveys, while one or more purse seine vessels will be chartered to capture fish for various purposes (e.g. identification of acoustic targets, disease studies, biological characteristics of spawning population). The experience provided by local fishermen in locating herring schools is essential to this project.

## PROJECT DESIGN

### A. Objectives

The overall goal of this project is to monitor the spawning population of Pacific herring in PWS to determine when this injured population has recovered. This project will also provide for development of a less costly but equally precise population assessment technique and a transition to ADF&G funding. The project has two specific objectives in FY98:

1. Estimate the biomass of spawning herring in PWS using SCUBA diving spawn deposition survey techniques such that the estimate is within  $\pm 25\%$  of the true value 95% of the time, and describe the age, sex and size composition of the spawning population.
2. Compare estimates obtained from spawn deposition and acoustic surveys and determine the best estimation technique to employ after FY98.

Estimation of the spawning herring population biomass, as well as its age, sex and size composition is the most important objective of this project.

## **B. Methods**

### **Objective 1: Spawn Deposition Survey and Biomass Estimation**

The survey design of the existing ADF&G spawn deposition project was modified for NRDA studies in 1989 to more accurately assess response of the PWS herring population to the *T/V Exxon Valdez* oil spill. Beginning in 1989, the spawn survey was conducted to obtain biomass estimates within  $\pm 25\%$  of the true biomass 95% of the time. Study design alterations included increasing the number of (1) SCUBA divers, (2) survey transects, and (3) skiff and diver surveys used to correct aerially mapped spawning area boundaries.

Biomass estimates based on spawn deposition surveys consist of three major components: (1) a spawn deposition survey; (2) age-weight-length (AWL), sex ratio, and fecundity sampling; and (3) egg loss determination.

***Spawn Deposition Survey Design.*** Survey design has been described in detail by Biggs and Funk (1988), and closely follows the two-stage sampling design of surveys used in British Columbia (Schwiebert et al. 1985) and Southeast Alaska (Blankenbeckler and Larson 1980, 1987). These surveys use random sampling for the first stage (transects) and systematic sampling for the second stage (quadrates within transects). Random sampling for the second stage is not feasible because of underwater logistical constraints (Schwiebert et al. 1985). Additionally, our surveys will be stratified by area to account for geographic differences and the potential of sampling discrete herring stocks. Areas surveyed may include Southeast, Northeast, North Shore, Naked Island and Montague Island depending upon the locations of spawning.

Mean egg densities along each transect will be combined to estimate an average egg density by area. Spawning bed width along each of the transects will be used to estimate average spawning bed width by area. Average width, average density, and total spawning bed shoreline length (from aerial surveys) will be used to estimate total number of eggs deposited in each summary area surveyed. Average fecundity and sex ratio, derived from AWL sampling, and estimates of total number of eggs deposited will be used to calculate herring population numbers and biomass. Based on variances obtained from the 1984, and 1988 to 1992 surveys, a minimum sampling goal of 0.035 % of all potential transects within the spawning area should ensure that the estimated biomass is within 25% of the true biomass 95% of the time. Based on the size of the sampling quadrat, there are 3,163 potential transects per kilometer. Therefore, 100 km of herring spawn would require 110 transects to meet our goals for accuracy and precision. Confidence intervals will be calculated assuming that total egg estimates follow a normal distribution.



***Spawn Deposition Survey Sampling Procedure.*** The general location of spawning activity will be determined from milt observed during scheduled aerial surveys that are part of an existing agency program. This information will be compiled and summarized on maps showing spawning locations and the number of days on which milt is observed. Total linear miles of shoreline containing herring spawn will be estimated from aerial survey maps and corrected by skiff and diver reconnaissance at the time of dive surveys. Skiff surveys will be performed close to shore at low tide by both walking along exposed intertidal areas and by viewing the shoreline from the skiff.

Each shoreline area containing herring spawn will be divided into the narrowest resolvable segments on the map scale (approximately 0.18 km). The total number of potential transects will be calculated from the total shoreline km of observed spawn. A minimum of 0.035% of all potential transects will be selected for dive surveys. Random numbers will be assigned to each potential transect and rounded to the nearest number divisible by 0.18 km to enable mapping of shoreline segments. Shoreline segments will be randomly selected and used to locate transects. Each transect selected will be assigned a sequential transect number and charted on waterproof field maps.

Diving on herring spawn will begin about 5 days after spawning has ceased to allow water turbidity due to milt to decrease and for the large numbers of sea lions usually present near spawning herring to disperse. Two three-person dive teams will complete the surveys. Each team will consist of a lead diver to count eggs (typically the person most experienced at this survey task), a second diver to record data, and a third diver on the surface performing as a tender. Diving and tending duties will be rotated daily. Based on information from previous PWS surveys, two diving teams can generally complete 6 to 12 transects daily under favorable weather conditions and in areas with average spawning density and distribution. A sample size total of 100 or more transects will require from 10 to 20 days of diving, depending upon weather and location of spawn. This time includes collection of diver calibration samples for a team of experienced divers. If inexperienced divers are hired, training will require about one additional week.

Location for each survey transect will be fixed as the dive skiff approaches the shore and before bottom profiles, bottom vegetation, or herring spawn are visible from the skiff. The tender will choose a shoreline feature to use as a reference point such as a tree, rock, or cliff located above the high tide line within the randomly selected shoreline segment. The sampling transect will extend seaward perpendicular to shore from this fixed reference point along a compass course.

Divers will estimate the numbers of eggs deposited within a sampling quadrat placed at regular intervals along the length of the transect. The sampling quadrat will consist of a 0.1 m<sup>2</sup> PVC pipe frame with a depth gauge and compass attached. The first quadrat location will be randomly selected within the first 5 meters of spawn. Succeeding quadrat locations will be systematically spaced every 5 meters along the compass course until the apparent end of the spawn is found. Within each quadrat, the lead diver will estimate the number of eggs in units of thousands (K) within the quadrat, communicating the numbers through hand signals to the

second diver to record. Number of eggs as well as vegetation type, percent cover, substrate, and depth will be recorded using a large weighted carpenter's pencil on water-proof plastic paper data forms attached to a clipboard. Divers will verify the end of the spawn by swimming at least an additional 20 m past the end of the spawn until a steep drop-off is encountered or vegetation is no longer present. Becker and Biggs (1992) documented methods used for diver surveys in greater detail including sample data forms, key codes for vegetation types, standard operating procedures for ADF&G diving, chemical recipes for sample preservatives, and other practical information.

Diver calibration samples will be collected throughout the dive survey and stratified by diver, vegetation type within four broad categories, and by egg density over three broad categories. Both divers will independently estimate the number of eggs on removable vegetation in each calibration quadrat. All egg-containing vegetation within the quadrat will be removed and placed in numbered mesh bags. The number of loose and attached eggs left after removal will be estimated by the lead diver and recorded. Based on accuracy estimated for previous survey results, approximately 90 calibration samples will be needed for each uncalibrated diver (less than three years survey participation) and 50 for each calibrated diver (three or more years survey participation). Calibration samples for each diver are to be taken from each of four vegetation categories: eelgrass (EEL), fucus (FUC), large brown kelp (LBK), and hair kelp (HRK); and from each of four ranges of egg densities: low (0-20,000), medium (20,000-80,000), and high (80,000-160,000), within each vegetation category. In 1996 the very high (> 160,000) category was added for eelgrass (EEL) and hair kelp (HRK) to better represent the density spread. Calibration samples will be preserved in Gilson's solution and labelled (Becker and Biggs 1992).

**Biomass Estimation.** Analysis of the spawn deposition survey data will be similar to methods used in 1988 (Biggs and Funk 1988). The biomass estimator will be

$$B = TB', \quad (1)$$

where

- B = estimated spawning biomass in tonnes,
- T = estimated total number of eggs (billions) deposited in an area, and
- B' = estimated tonnes of spawning biomass required to produce one billion eggs.

Estimates for T and B' will be derived from separate sampling programs and will be independent. The estimated variance for the product of the independent random variables T and B' will be (Goodman 1960)

$$Var(B) = T^2 Var(B') + B^2 Var(T) - Var(T) Var(B'), \quad (2)$$

where

Var(B') = an unbiased estimate of the variance of B', and

Var(T) = an unbiased estimate of the variance of T.

**Total Number of Eggs (T).** The total number of eggs deposited in an area will be estimated from a two-stage sampling program with random sampling at the primary stage, followed by systematic sampling at the secondary stage, using a sampling design similar to that described by Schwiebert et al. (1985). To compute variances based on systematic second stage samples, it will be assumed that eggs will be randomly distributed in spawning beds with respect to the 0.1 m<sup>2</sup> sampling unit. While this assumption will not be examined, in practice the variance component contributed by the second sampling stage will be much smaller than that contributed by the first stage, so violation of this assumption would have little effect on the overall variance. The total number of eggs (T), in billions, in an area will be estimated as

$$T = N\hat{y}10^{-6}/(1-R), \quad (3)$$

where

L = the shoreline length of the spawn-containing stratum in meters,

N = L/0.1<sup>0.5</sup> = the total number of possible transects,

0.1<sup>0.5</sup> = 0.3162 m = width of transect strip,

$\hat{y}$  = average estimated total number of eggs (thousands) per transect,

10<sup>-6</sup> = conversion from thousands to billions of eggs, and

R = estimated proportion of eggs disappearing from the study area from the time of spawning to the time of the survey.

Average total number of eggs per transect strip (in thousands) will be estimated as the mean of the total eggs (in thousands) for each transect strip using

$$\hat{y} = \frac{\sum_{i=1}^n \hat{y}_i}{n}, \quad (4)$$

$$\hat{y}_i = M\bar{y}_i, \quad (5)$$

where:

- n = number of transects actually sampled,
- i = transect number,
- $M_i = w_i/0.1^{0.5}$  = number of possible quadrates in transect i,
- $w_i$  = spawn patch width in meters measured as the distance along the transect between the first quadrate containing eggs and the last quadrate containing eggs, and
- $\bar{y}_i$  = average quadrate egg count in transect i (in thousands of eggs).

Average quadrate egg count within a transect,  $\bar{y}_i$ , will be computed as

$$\bar{y}_i = \frac{\sum_{j=1}^{m_i} y_{ij}}{m_i}, \quad (6)$$

where

- j = quadrate number within transect i,
- $m_i$  = number of quadrates actually sampled in transect i, and
- $y_{ij}$  = adjusted diver-estimated egg count (in thousands of eggs) from the diver calibration model for quadrate j in transect i.

The variance of T, ignoring the unknown variability in R, is similar to that given by Cochran (1963) for three stage sampling with primary units of equal size. In this case the expression is modified because the primary units (transects) do not contain equal numbers of secondary units (quadrates), and the variance term for the third stage comes from the regression model used in the diver calibration samples. Therefore the estimated variance of T, conditioned on R, is

$$Var(T) = \frac{[N^2(10^{-6})^2 \left[ \frac{(1-f_1)}{n} s_1^2 + \frac{f_1(1-f_2)}{\sum_{i=1}^n m_i} s_2^2 + \frac{f_1 f_2}{\sum_{i=1}^n m_i} s_3^2 \right]]}{(1-R)^2}, \quad (7)$$

where

$$s_1^2 = \frac{\sum_{i=1}^n (\hat{y}_i - \bar{y})^2}{n-1} = \quad (8)$$

variance among transects,

$$s_2^2 = \sum_{i=1}^n M_i^2 \sum_{j=1}^{m_i} \frac{(y_{ij} - \bar{y}_i)^2}{n(m_i - 1)} = \quad (9)$$

variance among quadrates,

$$s_3^2 = \sum_{i=1}^n \sum_{j=1}^{m_i} Var(y_{ij}) = \quad (10)$$

sum of the variances of the individual predicted quadrate egg counts from the diver calibration model,

$$f_1 = \frac{n}{N} = \quad (11)$$

proportion of possible transects sampled, and

$$f_2 = \frac{m_i}{M_i} = \quad (12)$$

proportion of quadrates sampled within transects (same for all transects).

**Diver Calibration.** Divers will be calibrated to correct systematic biases in their estimates of numbers of eggs. This calibration consists of the derivation of the relationship between diver estimates of eggs within a quadrate and actual counts obtained in the laboratory on the same eggs. Calibrations will be performed for each combination of diver and vegetation category as defined by the structural and phylogenetic similarities of egg-bearing plants. The four vegetation categories are designated eelgrass, fucus, hair kelp and large brown kelp (Becker and Biggs, 1992).

Diver bias will be determined using methods described in an as-yet unpublished report of the 1995 calibrations. The analysis will follow that described in the 1994 detailed project description in that the distribution of the random component will be assumed to be lognormal. However, the choice of random component (dependent vs. independent variable) will be reversed from that of previous analyses and diver estimate rather than laboratory egg count will be assumed lognormally distributed. Analysis of variance of Log(Diver Estimate), along with graphical methods, will be used to assess the significance of year, diver, and vegetation factors. The final model relating diver estimates to laboratory egg counts will be that which is simplest but retains suitable precision and lack of bias. Within the analysis of variance, attempts will be made to account for the repeated measures nature of the diver estimates, possibly using a split-plot analogy. Prediction of laboratory counts from the diver estimates made in the main spawn survey will, as a result of the designation of dependent and independent variables, be made in an inverse way. Variances of predicted laboratory counts will be estimated by the bootstrap method.

**Spawning Biomass per Billion Eggs (B').** Data from the herring sampling program for AWL, sex ratio, and fecundity will be used to estimate the relationship between spawning biomass and egg deposition. Once the age composition and sex ratio of a spawning population is determined, the average weight of the females in that population will be calculated. The relationship between fecundity and female weight will be used to calculate total numbers of eggs deposited and tonnes of herring spawners. The tonnes of spawning biomass required to produce one billion eggs (B') will be estimated as

$$B' = \frac{\bar{W}S}{F(\bar{W}_f)} 10^3, \quad (13)$$

where

- W = estimated average weight in grams of all herring (male and female) in the spawning population in an area,
- S = estimated ratio of total spawning biomass (male and female) to female spawning biomass,
- F(W<sub>f</sub>) = estimated fecundity at the average weight of females in the spawning population in an area, in numbers of eggs, and

$$\begin{array}{rcccl} 10^3 & = & \text{conversion factor} & = & \frac{10^{-6}}{10^{-9}} = \frac{\text{grams to tonnes}}{\text{eggs to billions}} \end{array}$$

Because average weight, sex ratio and fecundity will all be estimated from the same herring

samples. the estimates will not be independent. The variance of B' is approximately:

$$\begin{aligned}
 Var(B') = & (10^3)^2 \left( \left[ \frac{S}{F(\bar{W}_\rho)} \right]^2 Var(\bar{W}) \right. \\
 & + \left[ \frac{\bar{W}}{F(\bar{W}_\rho)} \right]^2 Var(S) \\
 & + \left[ \frac{\bar{W}S}{F(\bar{W}_\rho)^2} \right]^2 Var(F(\bar{W}_\rho)) \\
 & + 2Cov(\bar{W}, S) \left[ \frac{S}{F(\bar{W}_\rho)} \right] \left[ \frac{\bar{W}}{F(\bar{W}_\rho)} \right] \\
 & - 2Cov[\bar{W}, F(\bar{W}_\rho)] \left[ \frac{S}{F(\bar{W}_\rho)} \right] \left[ \frac{\bar{W}S}{F(F(\bar{W}_\rho))^2} \right] \\
 & \left. - 2Cov[S, F(\bar{W}_\rho)] \left[ \frac{\bar{W}}{F(\bar{W}_\rho)} \right] \left[ \frac{\bar{W}S}{F(\bar{W}_\rho)^2} \right] \right).
 \end{aligned} \tag{14}$$

Because S will be estimated from pooled or single AWL samples (depending on availability of fish), it will not be possible to estimate the covariance terms containing S, Cov(W, S) and Cov[S, F(W<sub>ρ</sub>)]. Because the term involving Cov[W, F(W<sub>ρ</sub>)] has been shown to be very small in previous analyses and probably contributes little to Var(B'), these covariance terms will not be included in the estimate of Var(B').

**Herring Age, Weight, Length, Sex Ratio, and Fecundity.** The largest portion of this project element has traditionally been part of an existing agency program conducted annually by ADF&G using volunteer commercial seine vessels to capture herring for basic biological sampling. AWL samples will also be collected from major concentrations of spawning herring using purse seine vessels under short term vessel charter in conjunction with acoustic surveys. Sampling will generally occur soon after concentrations of herring appear in nearshore areas and are accessible to purse seines. Samples will be taken periodically from major herring concentrations throughout PWS during the spawning migration. AWL samples collected during the peak of spawning in each summary area, as determined from aerial survey sightings of milt and herring schools, will be used to estimate age and sex composition as well as average herring size from all major biomass concentrations in each area.

AWL sampling will be stratified by date and area for test fishing catches in each spawning area. Sample size for each stratum will be set to simultaneously estimate proportions by age when sampling from a multinomial population (Thompson 1987). The goal will be to select the smallest sample size for a random sample from a multinomial population such that the probability will be at least  $1-\alpha$  (precision = 0.05) that all the estimated proportions will be simultaneously within 5% (accuracy = 0.05) of the true population age proportions. A sample size of 450 herring per stratum will be set to ensure that this level of precision and accuracy would be obtained for any number of age classes and proportions when less than 5% of the collected scales will be unreadable. Wilcock et al. (*In press*) provide a thorough description of PWS herring AWL sampling program procedures.

From an analysis of 5 years of fecundity data for PWS herring (personal communication, Tim Baker, Alaska Department of Fish and Game, Anchorage), Baker found that for a given year the relationships between herring weight and fecundity were very similar among areas, but less so among years for a given area. Year was found to be significant as were all interaction terms with year in an analysis of co-variance. As a result, we determined that it is probably important to collect fecundity data from PWS every year, but within a year, samples can be pooled across areas. Fecundity samples will be subsampled from all female herring in AWL samples and stratified by fish length. Egg and gonad weights will be measured and used to calculate average fecundity at the average female weight ( $F(\bar{W}_f)$ ).

A fecundity sampling goal was set such that fecundity estimates would contribute no more than 1% to the confidence interval width of the biomass estimate. This was achieved for surveys from 1988 through 1990 and 1992 during which area stratum sample sizes ranged from 100 to 400 fecundity samples and the standard error represented from 1.5 to 2.8% of the mean fecundity estimate. A sample size of 150 to 200 herring pooled across areas should be sufficient to maintain the coefficient of variation below 2.0%. To collect females over the range of possible sizes, we will sample 20 to 30 fish within each 10 mm length category from 181 to 250 mm standard length. In addition, we will collect 20 to 30 females 180 mm or smaller if available.

The female gonad weight will be assumed to be the equivalent of the weight of the ovaries removed from each female. Gonadal somatic index will be defined as the percentage of total herring weight represented by gonad weight and will be calculated by dividing the gonad weight by body weight of each fish sampled.

Mean weight and sex ratio will be estimated from AWL samples collected from each spawn deposition summary area. AWL samples collected during peak spawning in each area will be pooled to estimate mean weight and sex ratio for that area. Average weight and sex ratio for PWS will be estimated as a weighted average of estimates from all areas. Average weight and sex ratio for each area will be weighted by the escapement biomass estimate based on spawn deposition surveys for that area.

Sex ratio,  $S$ , will be calculated as the ratio of the number of herring of both sexes in AWL samples to the number of females. The binomial distribution is applicable to estimating the



proportion,  $p$ , of females in AWL samples, where  $S = 1/p$ . The variance of  $S$  is

$$Var(S) = \frac{S^2(S-1)}{n}, \quad (15)$$

where  $n$  is the number of fish in the AWL sample.

Average fecundity for PWS will be estimated from a fecundity-weight relationship as  $F(\bar{W}_f)$ , and used in equation 13 to estimate biomass from spawn deposition. The variance of estimated average fecundities will be approximated by the variance of predicted means from the fecundity-weight linear regression (Draper and Smith 1981)

$$Var[F(\bar{W}_f)] = s^2 \left[ \frac{1}{n} + \frac{1}{q} + \frac{(\bar{W}_f - \bar{WF})^2}{\sum (W_i - \bar{WF})^2} \right], \quad (16)$$

where

- $s^2$  = the residual mean square from the fecundity-weight linear regression,
- $\bar{W}_f$  = the average weight of female fish in the spawning population,
- $\bar{WF}$  = the average weight of females in the fecundity sample,
- $W_i$  = the weights of individual females in the fecundity sample,
- $n$  = the total number of females in the fecundity sample from each area, and
- $q$  = the total number of females in the representative AWL sample or pooled samples from the corresponding area.

A linear relationship between female body weight and fecundity will be used because Hourston et al. (1981) found that female body weight at spawning explained 70% of the variation in fecundity among individuals while length and age only explained another 2% of the variation.

A secondary purpose for determining average fecundity annually, will be to obtain information about natural fluctuations in reproductive potential in relation to fish size, fish growth, and environmental conditions. This information will be important for ecosystem studies such as project 98320 (SEA) that will test hypotheses about constraints to fishery production in PWS. For example, sea surface temperature appears to be an important natural factor affecting reproductive potential of herring. Tanasichuk and Ware (1987) found that sea surface temperatures 60 to 90 days before spawning best accounted for variations in size specific fecundity for herring in British Columbia, Canada. Using five years of PWS fecundity data, Biggs et al. (*in press*) showed egg production to be a function of fish body weight and to be

strongly correlated with sea surface temperatures 13 to 15 months prior to spawning. Egg weight was best correlated with sea surface temperatures 4 to 9 months prior to spawning and fecundity decreased as water temperatures increased.

## **Objective 2: Comparison of Spawn Deposition and Acoustic Biomass Estimates**

During 1997, preliminary comparisons of the precision, accuracy and cost of spawn deposition and acoustic biomass estimates will be made by survey area and year using data from 1995, 1996, and 1997. Paired spawn deposition and acoustic estimates are or will be available for one or more survey areas for each of these years.

The coefficient of variation will be used to compare the precision of spawn deposition and acoustic biomass estimates from each survey area. A bootstrap procedure will be used to obtain the distribution parameters for the coefficient of variation (Efron 1992). Bonferroni t-tests will be conducted to test for differences in the mean coefficient of variation between the two techniques by survey area (Kuehl 1994).

The accuracy of these biomass estimates cannot be directly determined, because we do not know the actual biomass of herring in each survey area. Therefore, we will indirectly evaluate the accuracy of the estimates obtained from acoustic and spawn deposition surveys by testing for differences among estimates obtained from several techniques. A scoring procedure will be used to track whether the estimates obtained from acoustic and spawn deposition surveys tend to be similar to estimates obtained from other techniques. Tests for differences between the acoustic and spawn deposition estimates by survey area will be conducted using a Bonferroni t-test (Kuehl 1994). The Friedman Rank Sums test will be used to test for differences among estimates obtained from aerial, spawn deposition and acoustic techniques by survey area (Hollander and Wolfe 1973). The Friedman Rank Sums test will also be used to test for differences among estimates obtained from the age-structured analysis (ASA) model, aerial surveys, spawn deposition surveys, and acoustics. These tests will be applied to estimates for the entire sound, because ASA model estimates are only available for the sound as a whole. The ASA model estimates are based primarily upon spawn deposition estimates from previous years and age-composition data (Funk 1995). Thus, inclusion of ASA estimates provides yet another source of information to evaluate accuracy. In each of the Friedman Rank Sums tests, year will be used as a block. A sign test will be used to explore planned comparisons if the rank sums tests indicates significant differences ( $\alpha < .05$ ) among estimates (Hollander and Wolfe 1973). Non-parametric techniques will be used, because variance estimates are not available for aerial survey or ASA model estimates.

A further assessment of the accuracy of spawn deposition and acoustic estimates will be made using a qualitative categorical variable indicating the relative accuracy (low, moderate, high) of the estimates obtained in each survey area. For the spawn deposition technique, this qualitative assessment will be based upon our ability to include all known spawn patches in the survey. Known spawn patches are not adequately surveyed in some years when high winds

preclude dive operations in exposed areas. Egg loss prior to dive surveys can be high in these cases (Rooper et al. 1996). For the acoustic technique, this assessment will be based upon our ability to include all known schools in the survey. Known schools of herring are not adequately surveyed in some years when the fish move into areas too shallow for boat operations (Thomas et al. 1996). Aerial survey and side-looking scanning sonar data will be used to evaluate the proportion of all herring schools in each area that have been included in the quantitative acoustic survey.

In FY98, comparisons of the precision, accuracy and cost of spawn deposition and acoustic biomass estimates will be made to determine the best estimation method to employ in future years. Data from 1995 through 1998 will be used in the analysis (Table 1). For each of the  $j$  cells in Table 1, a value will be assigned for precision, accuracy and unit cost. For precision, the value assigned to the  $j$ th cell will be the coefficient of variation for the respective estimation technique. For cost, the value assigned to the  $j$ th cell will be the total cost of obtaining the biomass estimate for the respective estimation technique including overhead. For accuracy, the value assigned to the  $j$ th cell will be a mean score ( $\bar{S}_j$ ) representing results from parametric tests, non-parametric tests, and qualitative assessments (Tables 2), i.e.

$$\bar{S}_j = \sum_i (S_{ij})/n_i \quad (17)$$

where  $S_{ij}$  is the score for the  $i$ th test in the  $j$ th cell and  $n_i$  is the number of tests (parametric, non-parametric, qualitative) for the  $j$ th cell.

A rank transformation will be performed on the values for precision, accuracy and cost, respectively, in each of the  $j$  cells referred to in Table 1. Analysis of variance will be used to test for differences in the rank-transformed cell values between the acoustic and spawn deposition techniques. Paired comparisons will also be conducted for precision, accuracy and cost, respectively, to test for differences in the rank-transformed cell values between the acoustic and spawn deposition techniques. The results from these analyses will be evaluated to determine the best estimation technique to employ after FY98.

Table 1: Survey areas and years for which data will be available to determine the best biomass estimation technique to employ after FY98. The numbers in the table are the  $j$  cell indices referred to in equation 25.

Year	Montague	Northeast	Southeast	PWS
1995	1			9
1996	2			10
1997	3	5	7	11
1998	4	6	8	12

Table 2: Example of the scoring procedure for results from parametric and non-parametric tests for differences in biomass estimates obtained from several estimation techniques for each of the  $j$  cells referred to in Table 1.  $S_i$  refers to the notation used for calculating mean scores in Equation 25. The example is for the acoustic estimation technique only. The scoring procedure for the qualitative assessment of accuracy is included for clarity. AC=acoustics, SD=spawn deposition, AR=aerial, ASA=age-structured analysis.

Type of Test	Estimates Not Diff. AC	Estimates Diff. AC	No. Estimates Diff. from AC	Score
Parametric ( $S_1$ )	AC	SD	1	.50
	AC, SD	-	0	1.00
Non-parametric test ( $S_2$ )	AC	SD AR	2	.33
	AC, SD	AR	1	.66
	AC, SD, AR	-	0	1.00
Non-parametric test ( $S_3$ )	AC	SD, AR, ASA	3	.25
	AC, SD	AR, ASA	2	.50
	AC, SD, AR	ASA	1	.75
	AC, SD, ASA	-	0	1.00
Qualitative ( $S_4$ )	-	-	-	.33
assessment	-	-	-	.66
	-	-	-	1.00

Several years of overlap in the use of acoustic and spawn deposition assessments is needed to adequately compare the two techniques and develop a link between the two biomass time series. At present, we have acoustic biomass estimates for herring spawning in the Montague survey area during 1995 and 1996. The 1995 acoustic estimate was similar to the spawn deposition estimate for the same area. However, the acoustic estimates from the 1996 surveys did not correspond well with the spawn deposition estimate for the same area. This was because many of the fish had already moved into shallow water to spawn at the time the acoustic survey was conducted. Several years of experience is needed to develop an adequate

understanding of the variations in weather conditions and fish behavior that affect the practicality of acoustic assessments on pre-spawning fish.

If we decide to transition to acoustic assessments, analysis of covariance and regression analysis will be used to develop a relationship between the two biomass time series. The analysis will include acoustic estimates as the dependent variable with spawn deposition estimates as a covariate and site and year as class variables. Survey area will be used as the sample unit in the analysis. This approach will model the relationship between the two biomass estimates and test for differences in the relationship by site and year. Such site and year differences may result from spatial and interannual variation in the timing and duration of spawning as well as the total number and distribution of spawners. Two additional years (97 & 98) of overlap in the use of acoustic and spawn deposition assessments is needed to test for site by year interactions in this analysis (Table 1).

### **C. Cooperating Agencies, Contracts and Other Agency Assistance**

Through a competitive bidding process, one or more purse seine vessels will be chartered to capture fish for AWL/fecundity samples, spawning adult herring for histopathology samples (project 98320S), and reproductive impairment samples for (project 98074). Depending upon the duration of the work and other competing uses, the ADF&G *R/V Montague* may be used as a sampling platform. In the event the *R/V Montague* is not available for use, another vessel will be secured on short term vessel charter agreement. This field work will occur over approximately 2 weeks during late March and early April.

One vessel will be chartered through a standard competitive bid process to be a research platform for spawn deposition surveys. This vessel will be used to house and transport SCUBA divers and their equipment. This portion of the project will last approximately 3 weeks from early to mid-April through early-May.

## SCHEDULE

### A. Measurable Project Tasks for FY 98 (Oct. 1, 1997 - Sept. 30, 1998)

September:	Finalize estimate of spawning biomass
November:	Finalize projection of 1998 run biomass
February:	1998 Biomass estimates - Dept. Forecast and Stock Assessment Reports
April:	<u>Submit FY97 annual report - biomass estimate</u> <u>Before onset of spawning:</u> Collect AWL, fecundity, disease, genetic stock ID, and bioenergetics samples <u>After onset of spawning:</u> Initiate dive surveys Submit FY99 Draft Detailed Project Description Complete dive surveys Begin lab processing of diver calibration and fecundity samples
June:	Complete calibration sample processing samples

### B. Project Milestones and Endpoints

The following milestones and endpoints will be achieved over the life of the project:

September 1997	Objectives 1-2:	Finalize estimate of spawning biomass of herring in 1997 using spawn deposition methodology.
September 1997	Objective 3:	Complete final report on herring recruitment model.
November 1997	Objective 3:	Compare spawn deposition and acoustic survey estimates of herring spawning biomass in 1997.
August 1998	Objectives 1-2:	Finalize estimate of spawning biomass of herring in 1998 using acoustic and spawn deposition methodology.
September 1998	Objective 2:	Compare spawn deposition and acoustic survey estimates of herring spawning biomass in 1998.
October 1998:	Objective 2:	Determine most appropriate and cost effective biomass estimation technique to employ in future years.
December 1998	Objective 1-4:	Complete project final report.

### C. Completion Date

No further Trustee Council funding will be requested for this project after FY98. However, monitoring of the abundance, age composition and size composition of the PWS Pacific herring spawning population will be continued beyond FY98 under ADF&G funding.

### PUBLICATIONS AND REPORTS

Scientific and technical aspects of the study will be subject to an internal peer review process within ADF&G's Commercial Fisheries Management and Development Division (CFMDD). Work plans, study design, and annual status reports will be subject to the peer review process established by the EVOS Trustee Council and Chief Scientist. Significant findings presented in annual and final reports will be submitted for publication in peer reviewed journals and presentation at scientific symposia as they are obtained. An final report will be submitted for FY98 by April 15, 1999.

### PROFESSIONAL CONFERENCES

Travel funds have been requested for this project to attend the EVOS annual workshop in Anchorage.

### NORMAL AGENCY MANAGEMENT

The following plan for transfer of herring biomass assessment back to normal ADF&G funding has been adopted (Table 3). After FY98, the results from this project will be used to decide whether to employ spawn deposition or hydroacoustic estimation techniques in future years.

Table 3: Planned transition of funding for herring biomass assessment program to ADF&G over the next three years. CDFU refers to the Cordova District Fishermen's Union.

Funding Source	FY96	FY97	FY98	FY99
EVOS TC	\$444.1	\$340.3	\$190.2	\$ 22.4
ADF&G	\$ 86.0	\$105.0	\$146.8	\$146.8
CDFU	\$ 40.0	?????	?????	?????

For FY98, the Trustee Council portion covers the spawn deposition survey, overhead costs, and most of the costs of project closeout. The ADF&G portion covers aerial surveys, test fishing, biological sampling, upgrade/purchase/maintenance of hydroacoustic equipment, continued training of personnel in hydroacoustics, and State vessel time. Again, the CDFU portion is not known since their grant has been exhausted. We intend to submit a request to the State Legislature for \$41,800 of test fishing receipt authority in FY98. This is the earliest such a request can be made due to the State Legislature's schedule. If approved, these test fishing monies will be used to fund acoustic surveys in FY98. However, test fishing can only be conducted if the herring biomass is above the productivity threshold.

In FY99, the Trustee Council portion covers final project closeout and overhead costs. The ADF&G portion covers aerial surveys, test fishing, biological sampling, upgrade/maintenance of hydroacoustic equipment, and State vessel time. Again, the CDFU portion is not known since their grant has been exhausted. In FY98, a request for test fishing receipt authority will be made to provide funding for the stock assessment methodology which has been identified as most appropriate and cost effective by this project. The dollar amount in Table 3 for FY98 may differ from that indicated depending on the assessment method chosen.

## **COORDINATION AND INTEGRATION OF RESTORATION EFFORT**

Project 98166 will be integrated closely with project 98320, SEA. Data management will be coordinated as outlined in SEA for integration of results. Other components of SEA will require sharing of information. Juvenile Herring Growth and Habitat Partitioning (98320T) will require location and abundance of spawn as well as information about age and size structure of sampled catches. Physical measurements taken for project 98166 may be useful to project 98320M. Information about spawn distribution will also be useful in drafting a study design for herring larval advection studies.

Project 98166 will also share information and resources with Project 98165, Herring Genetic Stock Identification in PWS. Additional samples required for this project beyond FY98 collections will be collected during AWL sampling and results will be used to refine our definition of stock structure. This improved stock definition will aid in recovery monitoring and the formulation of fisheries harvest strategies.

Other projects which will rely on sharing of resources with project 98166 for sample collection include Reproductive Impairment (98074), Somatic and Spawning Energetics of Herring/Pollock (98320U), and Disease Impacts on PWS Herring Populations (98162). The herring disease project (98162) relies on age-specific abundance estimates provided by Herring Natal Habitats to track changes in mortality associated with ichthyophonus and viral hemorrhagic septicemia.



Finally, integration of research will require data sharing and coordination with Project 98163, Forage Fish Influence on Injured Species. Herring are an important forage fish species. Herring and other forage fish are predators, competitors, and prey for each other at various stages throughout their life histories. Understanding the population dynamics of all forage species will lead to a better understanding of food availability, population fluctuations, and breeding success of birds and mammals that prey on them.

## **EXPLANATION OF CHANGES IN CONTINUING PROJECTS**

The field work associated with the egg loss study component of this project has been completed by the University of Alaska. A final report for this work has been submitted and will be included in the FY96 annual report. Funds are requested to complete the recruitment modelling study initiated by the University of Alaska under this project in FY95.

## **PRINCIPAL INVESTIGATOR**

Mark Willette  
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## **PERSONNEL**

**Mark Willette** (Principal Investigator), Alaska Department of Fish and Game, Commercial Fisheries Management and Development Division, P.O. Box 669, Cordova, Alaska 99574, (907)424-3214. **Education:** 1985 Master of Science, Fisheries Oceanography, University of Alaska Fairbanks. 1983 Bachelor of Science, Fisheries Science, University of Alaska Fairbanks. **Professional Experience:** March 1991 - present: Area Biologist with the Alaska Department of Fish and Game, Commercial Fisheries Management and Development Division in Cordova, Alaska. Supervised by Dr. Stephen Fried. Conduct various fisheries enhancement and evaluation projects in PWS including juvenile salmon growth studies, lake stocking, limnological investigations of sockeye salmon producing lakes, and quality control of coded-wire tagging at private hatcheries. Conduct fisheries oceanographic studies in PWS in cooperation with private hatcheries and University of Alaska investigators. Chairman of PWS Regional Planning Team. March 1986 - February 1991: Fisheries Instructor/ Assistant Research Professor, University of Alaska Fairbanks, School of Fisheries & Ocean Sciences, Supervised by Dr. Don Kramer. Conduct research on the effects of oceanographic conditions on the growth and survival of juvenile salmon in PWS, fish bioenergetics in an arctic lagoon ecosystem, age and growth of juvenile fish in the Chukchi and Bering Seas, ocean temperature

variability in the North Pacific Ocean and effects on pink salmon production, salmon feeding on the high seas. Design and implement a program of education, research, and public service to promote fisheries development in northwest Alaska. Teach college level course in oceanography. Teach a marine safety and vocational training courses in fisheries. **Research Projects:** Principal Investigator, Otolith Thermal Mass Marking of Hatchery Pink Salmon in Prince William Sound, 1995; Principal Investigator, SEA: Salmon Growth and Mortality, 1994-1995; Principal Investigator SEA: Salmon Predation, 1994-1995; Principal Investigator, Coghill Lake Sockeye Salmon Restoration, 1994-1995; Principal Investigator, Forage Fish Influence on Recovery of Injured Species - Fish Diet Overlap, 1994; Principal Investigator, Fish/Shellfish Study No. 4A, Early Marine Salmon Injury Assessment in Prince William Sound, 1991-1993; Co-investigator, Conceptual Model of the Ecosystem of Kasegaluk Lagoon, Alaska, 1989-1990; Co-investigator, Distribution, Abundance, Age and Growth of Fishes in the Southeast Chukchi Sea and Kotzebue Sound, 1987-1988.

#### **Selected Publications:**

Willette, T.M. 1996. Impacts of the Exxon Valdez Oil Spill on the migration, growth, and survival of juvenile pink salmon in Prince William Sound. *In* Proceedings of the Exxon Valdez Oil Spill Symposium, American Fisheries Society Symposium Series 18: 533-550.

R.T. Cooney, T.M. Willette, and S. Sharr. 1992. The effect of climate on Pacific salmon production in the northern Gulf of Alaska: examining the details of a natural experiment. *In* Proceedings of the International Symposium on Climate Change and Northern Fish Populations, Can. Spec. Publ. Fish. Aquat. Sci.

Willette, T.M. and R.T. Cooney. 1991. An empirical orthogonal functions analysis of sea surface temperature anomalies in the North Pacific Ocean and cross-correlations with pink salmon (*Oncorhynchus gorbuscha*) returns to southern Alaska. *In* Proceedings of the 1991 Pink and Chum Salmon Workshop.

Eggers, D.M., L.R. Peltz, B.G. Bue, and T.M. Willette. 1991. Trends in the abundance of hatchery and wild stocks of pink salmon in Cook Inlet, Prince William Sound, and Kodiak, Alaska. *In* Proceedings of the International Symposium on the Biological Interactions of Enhanced Salmonids, Can. Spec. Publ. Fish. Aquat. Sci.

**Member:** American Fisheries Society, Alaska Chapter.

**Greg S. Carpenter** (Co-investigator), Herring Fisheries Research Biologist, Alaska Department of Fish and Game, P.O. Box 669, Cordova, Alaska 99574. **Education:** Bachelors of Science, Wildlife Management, University of Alaska-Fairbanks, 1988. **Professional Experience:** October 1995 - present: Biologist with the Alaska Department of Fish and Game, Commercial Fisheries Management and Development Division in Cordova, Alaska. Supervised by Mark Willette. April 1989 - October 1995 Fishery Biologist I with the Alaska Department of Fish and Game, Commercial Fisheries Management and Development

Division in Cordova, Alaska. Supervised by Mark Willette. Assist with various fisheries enhancement and evaluation projects in PWS including juvenile salmon growth studies, lake stocking, limnological investigations of sockeye salmon producing lakes, and quality control of coded-wire tagging at private hatcheries. **Research Projects:** SEA: Salmon Growth and Mortality, 1994-1995; SEA: Salmon Predation, 1994-1995; Coghill Lake Sockeye Salmon Restoration, 1994-1995; Survey and Evaluation of Instream Habitat and Stock Restoration Techniques for Wild Pink and Chum Salmon, 1991-1993; Fish\Shellfish Study No. 4A, Early Marine Salmon Injury Assessment in Prince William Sound, 1991-1993.

### **Selected Publications:**

Willette, T. M., G.S. Carpenter, and E. Debevec. 1996. Herring spawn deposition and reproductive impairment, *Exxon Valdez* Oil Spill Restoration Project Annual Report (Restoration Project 95166), Alaska Department of Fish and Game, Cordova, Alaska.

Willette, T.M., G. Carpenter, S. Carlson, G. Kyle. 1995. Restoration of the Coghill Lake sockeye salmon stock, *Exxon Valdez* Oil Spill Restoration Project Final Report (Restoration Project 93024), Alaska Department of Fish and Game, Cordova, Alaska.

Willette, T.M., G. Carpenter, and E. Debevec. 1995. Sound Ecosystem Assessment: Salmon Growth and Mortality, *Exxon Valdez* Oil Spill Restoration Project Final Report (Restoration Project 94320A), Alaska Department of Fish and Game, Cordova, Alaska.

Edmundson, J.M., G.B. Kyle, T.M. Willette, and G.S. Carpenter. 1993. Juvenile sockeye salmon (*Oncorhynchus nerka*) stocking into Pass and Esther Pass Lakes, and nutrient enrichment of Pass Lake: and experiment on changes of the forage base. Fisheries Rehabilitation Enhancement and Development Report Series Number 125.

**Karen Hyer**, Biometrician, Alaska Department of Fish and Game, P.O. Box 669, Cordova, Alaska 99574. **Education:** Master of Science, Statistics, Oregon State University, 1995. Bachelor of Science, Marine Biology, Humboldt State University, 1987. **Professional Experience:** April 1996 - present: Biometrician, Alaska Department of Fish and Game, Commercial Fisheries Management and Development Division, Cordova, Alaska. Supervised by Brian Bue. Collaborate with biologist to develop analytic techniques for various fisheries evaluation projects in PWS. September 1993 - December 1995: Graduate Research Assistant, Oregon State University, Department of Statistics, Corvallis, Oregon. Supervised by Dr. Scott Urquhart. Design and perform statistical research and analysis for environmental and fisheries management programs. June 1995 - August 1995: Research Statistician, Oregon Health Sciences University, Center for Research on Occupational and Environmental Toxicology, Portland, Oregon. Supervised by Dr. Kent Anger. Collaborate with scientists to develop analytic techniques for identifying neurotoxic disorders associated with chemical exposure. May 1988 - September 1993: Fishery Biologist I / Fishery Biologist II, Alaska

Department of Fish and Game, Commercial Fisheries Management and Development Division, Anchorage, Alaska. Supervised by Dave Mesiar. Project leader for the Kuskokwim River sonar project. Design and implement experiments to evaluate the feasibility of using sonar to count migrating salmon. Develop a test-net sampling program for purpose of species apportionment. Research Projects: EMAP: Techniques for estimating river miles in the Midappalachian Highlands 1994-1995; Oregon State University: Predictors of Mercury Levels in Fish 1994; Oregon Health Science University: Socioeconomic factors affecting performance on behavioral tests 1995.

#### **Selected Publications:**

Hyer, K.E. et al. 1996. Kuskokwim River Sonar Progress Report, 1992. Alaska Department of Fish and Game, Commercial Fisheries Management and Development Division, Regional Information Report No. 3A96-24, Anchorage.

Hyer, K.E. et al. 1996. Kuskokwim River Sonar Progress Report, 1991. Alaska Department of Fish and Game, Commercial Fisheries Management and Development Division, Regional Information Report No. 3A96-25, Anchorage.

Hyer, K.E., D.C. Mesiar, and D.C. Huttunen, 1990. Kuskokwim River Sonar Feasibility Report, 1988. Alaska Department of Fish and Game, Commercial Fisheries Management and Development Division, Regional Information Report No. 3A90-11, Anchorage.

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# 1998 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1997 - September 30, 1998

Budget Category:	Authorized FFY 1997	Proposed FFY 1998						
Personnel	\$156.8	\$128.1						
Travel	\$2.6	\$0.6						
Contractual	\$63.8	\$33.8						
Commodities	\$8.3	\$5.6						
Equipment	\$1.2	\$0.0	LONG RANGE FUNDING REQUIREMENTS					
Subtotal	\$232.7	\$168.1	Estimated FFY 1999	Estimated FFY 2000	Estimated FFY 2001	Estimated FFY 2002		
General Administration	\$28.0	\$21.6						
Project Total	\$260.7	\$189.7	\$22.4					
Full-time Equivalents (FTE)	2.5	2.0						
			Dollar amounts are shown in thousands of dollars.					
Other Resources								
Comments:								
<p>Sea duty costs for each staff member are itemized under 'Overtime'.</p> <p>After FY98, a decision will be made to continue either spawn deposition or hydroacoustic biomass assessments at a reduced cost.</p>								

1998

Project Number: 98166  
 Project Title: Herring Natal Habitats  
 Agency: AK Dept. of Fish & Game

FORM 3A  
 AGENCY  
 PROJECT  
 DETAIL

Prepared:

1 of 4

4/10/97



**1998 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET**  
October 1, 1997 - September 30, 1998

Personnel Costs:			GS/Range/	Months	Monthly		Proposed
PM	Name	Position Description	Step	Budgeted	Costs	Overtime	FFY 1997
	M. Willette	Fishery Biologist III	18D	3.0	5,866	0	17.6
	G. Carpenter	Fishery Biologist II	16C	9.0	5,093	6,187	52.0
	B. Haley	Fish & Wildlife Technician III	11C	2.0	3,643	4,850	12.1
	C. Becker	Fish & Wildlife Technician III	11A	2.0	3,643	3,677	11.0
	M. Miller	Fish & Wildlife Technician III	11B	1.0	3,643	3,677	7.3
	K. Hyer	Biometrician I	17A	4.0	4,753	0	19.0
	S. Shipley	Fish & Wildlife Technician II	9A	1.0	3,229	0	3.2
	A. DiBicarri	Fish & Wildlife Technician I	7A	1.0	2,696	0	2.7
	P. Trautman	Field Office Assistant	9A	1.0	3,200	0	3.2
Subtotal				24.0	35,766	18,391	
Those costs associated with program management should be indicated by placement of an *.							Personnel Total \$128.1
Travel Costs:			Ticket	Round	Total	Daily	Proposed
PM		Description	Price	Trips	Days	Per Diem	FFY 1996
		RT Cordova-Anch., Attend EVOS annual workshop, 1 staff	200	1	4	95	0.0
							0.0
							0.6
							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 \$0.6

**1998**

Project Number: 98166  
Project Title: Herring Natal Habitats  
Agency: AK Dept. of Fish & Game

FORM 3B  
Personnel  
& Travel  
DETAIL

# 1998 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1997 - September 30, 1998

Contractual Costs:		Proposed
Description		FFY 1997
Publication costs		1.0
CPR /FIRST AID training (4 classes @ \$100 per class)		0.4
Vessel charter (18 days @ \$1500 per day; logistical support for dive crew)		27.0
Dive physicals		2.5
Network operation & maintenance		0.5
Aircraft charter (3 hours @ \$275 per hour)		0.9
Dive equipment repair & maintenance		1.5
When a non-trustee organization is used, the form 4A is required.		
<b>Contractual Total</b>		<b>\$33.8</b>
Commodities Costs:		Proposed
Description		FFY 1997
Office/Lab supplies		1.0
Dive gear replacement parts		1.0
Groceries for (7 people x 18 days x \$20 per day)		2.5
Skiff fuel (240 gals @ \$1.67 per gal.)		0.4
Misc. field sampling supplies (outboard oil, spark plugs, bilge pumps, sample frame parts, etc.)		0.7
<b>Commodities Total</b>		<b>\$5.6</b>

**1998**

Project Number: 98166  
Project Title: Herring Natal Habitats  
Agency: AK Dept. of Fish & Game

FORM 3B  
Contractual &  
Commodities  
DETAIL

**1998 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET**  
 October 1, 1997 - September 30, 1998

New Equipment Purchases:		Number of Units	Unit Price	Proposed FFY 1997
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.			<b>New Equipment Total</b>	\$0.0
Existing Equipment Usage: (partial inventory)		Number of Units	Inventory Agency	
Description				
Wave and tide recorder with computer interface		3	ADFG	
Alumaweld Sea Dory (20 ft)		1		
Boston Whaler (17 ft)		1		
Dive regulators		29		
Dive depth\pressure gauges		14		
Dive backpacks		9		
Dive jumpsuits (various sizes)		12		
Dry suits (various sizes)		14		
Dive tanks		21		
(complete inventory on file at ADFG, Cordova)				

**1998**

Project Number: 98166  
 Project Title: Herring Natal Habitats  
 Agency: AK Dept. of Fish & Game

FORM 3B  
 Equipment  
 DETAIL

98169-BAA

# **A Genetic Study to Aid in Restoration of Murres, Guillemots and Murrelets to the Gulf of Alaska; Submitted Under the BAA**

Project Number: 98-169  
Restoration Category: Research  
Proposer: Queen's University (V.L. Friesen) & DOI (J.F. Piatt)  
Lead Trustee Agency: DOI  
Cooperating Agencies: Alaska SeaLife Center:  
Duration: 2nd year, 4-year project  
Cost FY98: \$84.8  
Cost FY99: \$86.2  
Cost FY00: \$13.8  
Geographic Area: Gulf of Alaska and neighboring areas  
Injured Resource: common murre, pigeon guillemot, marbled and Kittlitz's murrelets

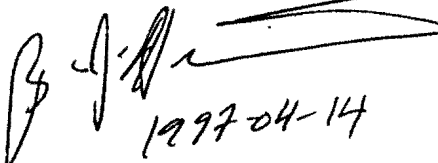
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APR 15 1997

**EXXON VALDEZ OIL SPILL  
TRUSTEE COUNCIL**

## **ABSTRACT**

Populations of common murres, pigeon guillemots, and marbled and Kittlitz's murrelets from the Gulf of Alaska are failing to recover from the *Exxon Valdez* Oil Spill. We propose to continue our genetic analyses to aid in their restoration by 1) determining the geographic limits of the populations affected by the Spill, 2) identifying sources and sinks, and 3) identifying appropriate reference or 'control' sites for monitoring. As incidental results, this study will also reveal cryptic species and subspecies, indicate the role of inbreeding and small effective population sizes in restricting recovery, and suggest suitable source colonies for translocations.

**B. J. HUTCHINSON**  
Director, Office of Research Services  
Queen's University, Kingston, Ontario

  
1997-04-14

## INTRODUCTION

Common murres (*Uria aalge*), pigeon guillemots (*Cepphus columba*), marbled murrelets (*Brachyramphus marmoratus*) and Kittlitz's murrelets (*B. brevirostris*) appear not to be recovering from the the *Exxon Valdez* Oil Spill. An understanding of the genetic structure and dynamics of these populations is critical for their restoration. Although the term *population* commonly is used to refer to any group of organisms, it is defined biologically as a group of organisms that share a common gene pool due to interbreeding. A population may consist of several localized subpopulations, such as colonies of seabirds. If gene flow among colonies is low, either because levels of dispersal are low or because immigrants have low fitness, populations will comprise colonies; if dispersal is more widespread, then populations may include several colonies within a region. For example, colonies of thick-billed murres (*Uria lomvia*) within the North Atlantic appear to constitute a single panmictic population that is genetically isolated from colonies in the North Pacific (Birt-Friesen et al. 1992). Some species, such as many gulls and cormorants, appear to comprise 'metapopulations' - networks of colonies that disappear naturally and are recolonized by immigrants from other sites over time periods ranging from a few generations to tens of thousands of years. Generally, subpopulations of a metapopulation are geographically isolated but exchange migrants on either a regular or intermitant basis (Levins 1969). In some species, individual subpopulations may constitute 'sources' or 'sinks': subpopulations in optimal sites have high productivities and low mortalities, and act as net exporters or 'sources' of breeders for other sites; sink populations occur in suboptimal habitats and have low productivities, so require immigration to maintain their numbers (Pulliam 1994). For example, the colony of ancient murrelets (*Synthliboramphus antiquus*) on Reef Island may act as a source of immigrants maintaining the colony at Limestone Island, where predation by raccoons and rats is high (A.J. Gaston, pers. comm.). Results from theoretical models, as well as practical experience with other species, suggest that in long-lived animals with low productivities, even populations that appear healthy and stable may decline precipitously and disappear over a very few years due to demographic and genetic problems.

Information about the genetic structure and dynamics of populations of murres, murrelets and guillemots is needed for restoration for three main reasons.

*Definition of the geographic limits of the affected populations.*-Many seabirds killed by the *Exxon Valdez* Oil Spill were migrating: the 'affected' zone, or the populations that were affected by the Spill and require restoration effort, may be geographically distant from the actual Spill zone. Genetic data enable identification of breeding populations and thus the geographic limits of the populations of birds killed by the Spill. If colonies are essentially panmictic and/or constitute metapopulations, they should recover without assistance within a number of generations. For example, double-crested cormorants (*Phalacrocorax auritus*) have recolonized many sites from which they were extirpated by pesticides in the 1950-1960s. However, if colonies constitute numerous localized populations, they probably will not naturally recolonize sites affected by the Spill, and may require human assistance for recovery. For example, common murres have failed to repopulate colonies in southern Quebec from which they were extirpated by egging and shooting in the late 1800s and early 1900s (e.g. Tuck 1961). Clarification of the geographic limits of the affected populations is especially important because colonies appeared to be declining before the accident, and the relative importance of the Spill versus other

environmental effects (such as prey availability) on population decline and recovery is unclear.

*Identification of sources and sinks.*-Genetic data can provide measurements of rates of immigration into and emigration out of colonies, and thus enable identification of sources and sinks. If colonies affected by the Spill represent sources, then their restoration will be critical. For example, protection of the ancient murrelet colony on Reef Island may be essential to the longevity of the local population of murrelets. If a colony represents a sink, its restoration may be a waste of resources and may actually prevent recovery of the total population.

*Environmental monitoring.*-Genetic data enable identification of appropriate reference or 'control' sites from which to obtain baseline data for monitoring, restoration and modeling, e.g. to determine if a seabird colony has recovered 'normal' functioning. Demographic parameters may be very different for genetically divergent populations, even if they occur in ecologically similar or geographically proximate areas. For example, common murre breeding in Washington have different breeding chronologies from those at neighboring colonies in British Columbia; K. Warheit (Washington State Department of Fish and Wildlife) and V.L.F. are presently investigating their genetic relationships.

Two other types of information that are useful for conservation and restoration are produced incidentally by genetic studies.

*Population uniqueness and cryptic species.*-A colony's uniqueness (e.g. its endemism or genetic distinctiveness) may be used to prioritize restoration efforts. Most importantly, genetic data enable the identification of cryptic species - populations that are similar in appearance but that represent separate, non-interbreeding species. For example, genetic comparisons revealed that North American and Asiatic subspecies of the marbled murrelet actually represent reproductively distinct species that have been genetically isolated for 5-6 million years (Friesen et al. 1996a). These two taxa therefore must be managed independently. Preliminary data also suggest that Kittlitz's murrelets from Kachemak Bay are highly divergent from those from Attu Island, and that the two populations may represent cryptic subspecies or species.

*Small effective population size and inbreeding.*-The *effective size* of a population is the number of individuals that actually contribute to the gene pool of the population, and may be one or two orders of magnitude lower than the census size due to unequal breeding success and population bottlenecks. For example, the North Atlantic population of thick-billed murre consists of approximately 2.5 million breeding pairs (Nettleship and Evans 1985), but appears to have a long-term effective size of only ~15,000 females (Friesen et al. 1996b). As a population's effective size declines, its genetic resources become depleted (Allendorf and Leary 1986, Gilpin and Soulé 1986). Initially this depletion involves loss of rare variants (alleles) from the population, but ultimately it includes loss of individual variation (heterozygosity) due to increased inbreeding. Low heterozygosity often is associated with low fitness due to a decrease in such factors as survival, growth, reproductive success and disease resistance (e.g. O'Brien & Evermann 1988, Vrijenhoek 1994). For example, low reproductive success of cheetahs has been attributed to low genetic variation resulting from population bottlenecks and inbreeding (O'Brien & Evermann 1988). Several lines of evidence suggest that if a population declines below an effective size of approximately 50 individuals, it may enter an extinction vortex in which

inbreeding, deleterious alleles and stochastic effects combine synergistically to accelerate extinction (Gilpin and Soulé 1986). Genetic information may be used to estimate effective population size (Nei and Li 1979), and thus to determine the extent to which small effective population sizes and inbreeding are inhibiting population recovery.

*Translocations.*-If breeding success within a colony is low due to inbreeding depression, translocation of small numbers of individuals from other sites may be desirable. Translocations may also be useful for boosting recruitment. Ideally, sources of animals for such introductions should be neighboring colonies within the same population or a closely related population. Genetic data are important for determining which colonies are genetically appropriate sources to prevent both inbreeding and introductions of genetically incompatible or inappropriate individuals. For example, after a captive breeding program was designed to restore the dusky seaside sparrow (*Ammodramus maritimus nigresens*) by hybridizing the last remaining males with females of the morphologically similar Scott's seaside sparrow (*A. m. peninsulae*), genetic analyses indicated that Scott's seaside sparrow was not the most closely related subspecies to the dusky seaside sparrow and therefore not the most appropriate choice for captive breeding (Avisé and Nelson 1989).

### **Methodology**

Generation of the population genetic information necessary for restoration of most species of birds breeding at high latitudes, such as the seabirds affected by the Spill, requires highly sensitive molecular markers. Although gene flow and population genetic structure can be approximated from demographics (e.g. Rockwell and Barrowclough 1987), generation of these data involves long-term banding studies and is extremely labour-intensive, especially for species with secretive nesting habits, such as marbled and Kittlitz's murrelets. Furthermore, estimates of genetic divergence from demographic data tend to miss occasional mass migrations, which may be important sources of gene flow in seabirds (e.g. Nettleship and Evans 1985). Traditional molecular methods such as protein electrophoresis also are not suitable for measuring genetic subdivision in populations that breed at high latitudes due to low levels of variability (Evans 1987). Although DNA fingerprinting can reveal high levels of variability, it is expensive, laborious and time-consuming, and exhibits levels of homoplasy (genetic 'noise') too high for comparisons of populations.

Recent innovations in molecular genetics, especially the polymerase chain reaction (PCR, or DNA amplification) provide several advantages over previous methods of genetic analysis. Most importantly for the present purposes, they enable DNA sequences to be compared directly among individuals from different populations (e.g. Kocher et al. 1989, Birt-Friesen et al. 1992, Quinn 1992, Wenink et al. 1994). Furthermore, they allow researchers to focus their attention on genes with high levels of variability, such as mitochondrial DNA (mtDNA), nuclear introns or microsatellites. Unfortunately, most existing PCR-based protocols either require extensive groundwork (e.g. analysis of microsatellite loci), or are expensive and laborious (e.g. analysis of sequence variation in mtDNA); however, most approaches may be combined with various mutation-detection methods, such as the analysis of single-stranded conformational polymorphisms (SSCPs) and denaturing gradient gel electrophoresis (DGGE; Lessa & Applebaum 1993; Friesen et al. 1996, in press) to reduce both the cost and time involved in population-level surveys. In the present study, we propose to use a combination of analyses of



the mitochondrial control region, microsatellite loci, and nuclear introns. Previous experience indicates that this combination of techniques is optimal for assaying genetic variation within and among neighboring colonies of seabirds (see below).

### ***Past efforts and results***

Previous studies of relatively slowly evolving genes (allozymes and the mitochondrial cytochrome *b* gene) indicated that Atlantic and Pacific populations of both common and thick-billed murres are genetically distinct (Birt-Friesen et al. 1992, Friesen et al. 1996b).

Differentiation among Atlantic populations of thick-billed murres appears to be weak, but Atlantic colonies of common murres exhibit clinal variation in cytochrome *b* genotype frequencies. Sample sizes within the Pacific were very small and restricted to the Bering Sea, but preliminary results indicated that colonies differ in allele frequencies, and thus may represent genetically isolated populations (V.L.F. unpubl. data). Previous studies of geographic variation in marbled murrelets using allozymes and cytochrome *b* indicated that the Asian and North American subspecies represent cryptic species that have been genetically isolated for 5-6 million years and that must be managed independently (Friesen et al. 1996a). Preliminary results of this study also indicated significant differentiation among North American populations of murrelets, suggesting that they do not represent a single population. Furthermore, preliminary analysis of small numbers of Kittlitz's murrelets from Kachemak Bay and Attu Island revealed that these populations are highly divergent and may represent cryptic subspecies or even species: further genetic analyses are critical for the conservation of this species. In all these studies, levels of variability in proteins and the cytochrome *b* gene were too low for comparisons of neighboring colonies.

V.L.F. and members of her research group have now developed protocols for assaying variation in more rapidly evolving genes, including introns of nuclear genes (e.g. genes of the major histocompatibility complex, which are involved in disease resistance), the mitochondrial control region (which evolves up to ten times faster than cytochrome *b*), and microsatellite loci, and are applying them to conservation studies of murres and murrelets from throughout the North Pacific. Results to date confirm the genetic distinctiveness of Asian and North American forms of the marbled murrelet, as well as the existence of genetic differences among local populations of murrelets. In addition, we have completed surveys of variation in the mitochondrial control region (a notoriously difficult gene to analyze in birds) among populations of guillemots from throughout the Northern Hemisphere (Kidd and Friesen submitted), and have found colony-specific differences. We have also begun a survey of control region variation within a colony of thick-billed murres (Ibarguchi et al., in progress), and are developing protocols for analysis of microsatellite loci in murres (Ibarguchi et al., unpubl.), murrelets (Congdon et al., in progress) and guillemots (Crossman and Friesen, in progress).

In February 1997, we received funding from the *EVOS* Trustee Council to analyze variation in mitochondrial DNA, microsatellites and introns among available samples from murres, guillemots and murrelets from the area of the *Exxon Valdez* oil spill and neighboring sites. To date we have been refining protocols for analyses of microsatellites and control regions from these birds, and have begun screening samples for variation in several introns. Preliminary results suggest that murres from Kachemak Bay and Chisik Island differ both in allele frequencies and levels of genetic variability from those from the Barren Islands.

### ***Proposed project***

In the present project, we propose to continue our genetic analysis of common murres, pigeon guillemots, and marbled and Kittlitz's murrelets. Specifically, we proposed to screen samples of all four species for variation in the mitochondrial control region, microsatellite loci, and nuclear introns. This work is necessary 1) to determine the geographic limits of the affected populations, 2) to identify sources and sinks, and 3) to identify appropriate reference or 'control' sites for monitoring. As incidental results, this study will also identify cryptic species and subspecies, indicate the role of inbreeding and small effective population sizes in restricting recovery of the target species, and suggest appropriate source populations for translocations (if necessary). In FY98 we will screen samples collected in FY97 from within the Spill area and neighboring sites; in FY99, our analyses will be expanded to include samples from more distant sites.

## **NEED FOR THE PROJECT**

### **A. Statement of Problem**

Seabirds of the family Alcidae (auks, guillemots, murrelets, auklets and puffins) are highly vulnerable to marine oil pollution due to the large amount of time they spend resting on the ocean surface, as well as their dependence on marine fish and invertebrates for food. Many species of alcids suffered heavy mortality associated with the *Exxon Valdez* Spill; for example, the estimated mortality for common murres was in the hundred of thousands. Although guillemots and murrelets were declining prior to the Spill, the accident probably increased their rate of decline. Numbers of common murres, pigeon guillemots and marbled murrelets apparently are not recovering; the state of recovery of Kittlitz's murrelets is unknown. The reasons for the failure of these species to recover (as well as for the prespill declines) are unclear, but may be due to availability and quality of prey (currently being investigated through the APEX Predator Experiment and Nearshore Vertebrate Predator Project), and/or genetic problems such as genetic isolation of colonies or inbreeding. We propose to use state-of-the-art genetic techniques to aid in the restoration of these species.

### **B. Rationale/Link to Restoration**

The proposed investigation will aid recovery in three main ways:

- 1) It will provide a measure of the geographic extent of the populations of murres, murrelets and guillemots affected by the Spill. This will enable determination of the 'affected' zone, and the relative importance of the Spill versus other environmental effects on the decline and recovery of different colonies. It will also enable determination of the extent to which a lack of natural gene flow is inhibiting recovery, and thus the need for active restoration.
- 2) By providing measures of immigration into and emigration out of colonies, it will identify sources and sinks. This will help to direct restoration efforts to the most productive colonies.
- 3) The study will help to identify appropriate reference or 'control' sites for monitoring, by delineating the geographic limits of populations.

Three additional, incidental benefits will be derived from the study:

- 1) It will identify cryptic species or subspecies, an especially likely possibility for Kittlitz's

murrelets. This will help to prioritize restoration efforts.

- 2) By providing a measure of genetic variability and inbreeding within populations, it will indicate the role of inbreeding depression and small effective population sizes in restricting recovery of the target species. This will reveal the importance of reintroductions for restoration.
- 3) It will indicate suitable source populations if reintroductions are necessary either to supplement numbers or to reduce inbreeding depression.

### **C. Location**

This project will require collection of blood, feather and/or tissue samples from birds breeding throughout the Pacific basin, mostly in Alaska (Table 1). As much as possible, tissue will be obtained from museum specimens, and blood and blood feathers ('pin' or growing feathers) will be obtained from chicks or adults during banding. Birds being collected for ongoing diet studies in Alaska (J.F.P.) also will be used as a source of tissue. In year 1 (FY97), samples that are already in hand from the Spill area and immediately adjacent sites are being analyzed. In year 2 of the project (FY98), emphasis will be placed on obtaining additional samples from key sites within the Spill area, as well as from neighboring areas in Alaska (Gulf of Alaska, Bering Sea, Aleutian Islands, Chukchi Sea). In year 3 (FY99), sampling efforts will be expanded to include the Sea of Okhotsk, Japan, and the southern United States, using assistance from colleagues in those areas. Results of the project will aid in the restoration of populations of murres, murrelets and guillemots to areas affected by the Spill.

## **COMMUNITY INVOLVEMENT**

Unfortunately, the bulk of work involved in the proposed project must be conducted by highly trained personnel in a specially equipped research laboratory. If available, a local student interested in graduate work in conservation genetics may be hired by V.L.F. We will attempt to obtain tissue samples from seabirds harvested for subsistence purposes when possible. Sample collections may require chartering local vessels and paying for assistance from local experts, hunters or vessel operators (see **Methods**). Information about the age of colonies, which is needed for interpretation of genetic results, will be sought from traditional knowledge. Project objectives and interim results will be communicated to local residents through popular reports in the Trustee Council newsletter.

## **PROJECT DESIGN**

### **A. Objectives**

The primary purpose of this project is to conduct a genetic analyses to aid in the restoration of common murres, pigeon guillemots, and marbled and Kittlitz's murrelets to areas affect by the *Exxon Valdez* Oil Spill. We have three main objectives for each species:

- 1) To determine the geographic extent of the populations affected by the Spill.

- 2) To identify source and sink colonies.
- 3) To identify appropriate reference or 'control' sites for monitoring.

As incidental results, we should also be able:

- 4) To identify cryptic species or subspecies.
- 5) To measure coefficients of inbreeding and effective population sizes.
- 6) To identify appropriate source populations for translocations, if necessary.

## **B. Methods**

In year 1 (FY97) we are analyzing genetic variation in samples of murres, guillemots and murrelets that are already in hand from the Spill area and immediately adjacent sites. In year 2 (FY98), we propose expand our initial surveys to include more sites from inside the Spill area and immediately adjacent sites. Analysis of these samples is necessary to define the geographic limits of the breeding populations. If necessary (if the geographic limits of the populations remain undetermined), samples from more distant sites will be collected and analyzed in year 3 (FY99).

*Common Murres.*- Comprehensive assessment of genetic differentiation and gene flow among populations of common murres may require analysis of samples from up to 15 sites (Table 1). Samples from California, Washington and Oregon are already being analyzed in conjunction with Dr. Ken Warheit (Washington State Department of Fish and Wildlife) as part of an independently-funded study.

*Marbled Murrelets.*- Much of the desired information for marbled murrelets (specifically, variation in nuclear introns) already is being collected by V.L.F. et al. under a contract from the Canadian government. Assessment of genetic markers for murrelets breeding within the Spill area will require collection and analysis of samples from additional sites (Table 1). Variation in the control region and microsatellite loci will have to be assayed for all samples.

*Kittlitz's Murrelets.*- Kittlitz's murrelets are rare and notoriously secretive in their nesting sites. Samples of these birds therefore are extremely difficult to obtain. We will analyze any samples that we can obtain from anywhere in their range either through collections for dietary analyses or from incidental catches in gill nets or oil slicks. We anticipate obtaining a total of approximately 50 samples.

*Pigeon Guillemots.*- A large-scale survey of genetic variation in guillemot mtDNA has already been conducted in V.L.F.'s laboratory (e.g. Kidd and Friesen submitted). Measurement of genetic differentiation and gene flow and identification of genetic markers for guillemots breeding within the Spill area will require analysis of variation in introns and microsatellite loci in at least 30 samples from each of 12 additional sites (Table 1).

Many of the necessary baseline samples already have been obtained opportunistically during

other research project through the assistance of Vern Byrd and Dave Roseneau (Alaska Maritime National Wildlife Refuge), Jay Pitocchelli, Tom van Pelt and Lindsey Hayes (National Biological Service, Anchorage), Alex Pritchard (University of Alaska), Jan Hodder (Oregon Institute of Marine Biology) and Kathy Martin (Canadian Wildlife Service). Other samples are available from tissue collections at the University of Alaska Museum and the Burke Museum (University of Washington). Funding is required in FY98 to complete collections of samples from the Spill area. To fill gaps in numbers and distributions, a concerted effort will be required to collect blood or specimens at key geographic sites (Table 1). This may require air travel to sites of interest, chartering local vessels, and paying for assistance from local experts, hunters or vessel operators. Permits for collection of seabirds are required from the U.S. Fish and Wildlife Service, the State of Alaska (ADF&G) and the Animal Care Committee of Queen's University, and will be obtained by J.F.P. and V.L.F. prior to collections.

Genetic analyses will be conducted by the technicians as samples become available. Previous work suggests that each person can process approximately 4500 samples per year. Analysis of 20 loci (2 mitochondrial genes [two parts of the control region], 8 microsatellite loci and 10 introns) for each of approximately 1200 samples (excluding 150 murrelets already being analyzed by V.L.F. et al.) is expected to require approximately 5.5 person-years. Screening 200 carcasses salvaged from the Spill for population-specific genetic markers will require approximately 0.5 person-years in year 3 (FY99). Data will be analyzed using standard methods developed for analysis of data from protein electrophoresis and sequencing (e.g. Swofford & Selander 1981; Nei 1987); inbreeding coefficients will be calculated as estimators of genetic variabilities; genetic differentiation will be calculated using Wright's *F* statistics and its analogues (e.g. Excoffier et al. 1992); historical gene flow among colonies will be estimated using Slatkin's (1985) method of private alleles and Hedrick's (1971, 1975) *U* statistic. Results of these analyses will indicate cryptic species and subspecies, sources and sinks, colony-specific markers for impact assessment, appropriate control sites for monitoring, and source populations for reintroductions. Finally, representatives of each allele detected in the population surveys will be sequenced to identify effects of selection (e.g. Abernethy 1994, Simonsen et al. 1995), to measure contemporary gene flow using the method of coalescence (Slatkin and Maddison 1989), and to estimate effective population sizes (Nei and Li 1979).

### **C. Cooperating Agencies, Contracts, and Other Agency Assistance**

Collections of blood and tissue will be coordinated with other agencies (museums, wildlife agencies, etc.) by V.L.F. and J.F.P. Vessel charters will be arranged with the Alaska Maritime National Wildlife Refuge and with private boat operators throughout the study area. No additional contracts or cooperating agencies are required to complete this project. Genetic analyses are enabled in part by previous support from the Natural Sciences and Engineering Research Council (Research Grant held by V.L.F) and the Environmental Innovations Program of Public Works and Government Services Canada (contract held by V.L.F and T.P. Birt).

## **SCHEDULE**

### **A. Measurable Project Tasks for FY 98**

Jan. 1 '98 - Jan. 30 '98:	Technicians screen samples available from FY97 from the Spill area and neighboring sites for variation in the mitochondrial control region
Jan. '98:	PIs attend Annual Restoration Workshop
Feb. 1 '98 - Jun. 30 '98:	Technicians screen samples from FY97 for variation at 8 microsatellite loci
Jan. 1 '98 - Apr. 30 '98:	PIs arrange logistics for sample collections
May 1 '97 - Aug. 30 '97:	Blood, feather and tissue samples collected from sites in Alaska by J.F.P.
Jul. '98:	VLF and/or JFP present interim results at conferences
Jul. 1 '98 - Dec. 31 '98:	Technicians screen samples from FY97 for variation at 10 nuclear introns

## **B. Project Milestones and Endpoints**

Jan. '97:	PIs attend Annual Restoration Workshop
Mar. 31 '97:	Technicians complete development of microsatellite protocols for guillemots; refine protocols for analysis of introns and control regions for each species as necessary
Aug. 31 '97:	Field collections for FY97 completed (some Alaskan sites)
Dec. 31 '97:	Technicians complete screening of samples available prior to FY97 for variation in the mitochondrial control region, 8 microsatellite loci and 10 introns
Jan. '98:	PIs attend Annual Restoration Workshop
Apr. 15 '98:	VLF completes annual report for FY 97
Aug. 31 '98:	Field collections for FY98 completed (Alaskan sites)
Dec. 31 '98:	Technicians complete screening of samples available from FY97
Jan. '99:	PIs attend Annual Restoration Workshop
Apr. 15 '99:	VLF completes annual report for FY98
Aug. 31 '99:	Field collections for '99 completed (sites outside Alaska)
Dec. 31 '99:	Technicians complete screening of all samples for variation
Jan. '00:	PIs attend Annual Restoration Workshop
Apr. 15 '00:	VLF completes annual report for FY99
Jun. 30 '00:	VLF and technicians complete data analysis ( <u>including all analyses outlined in Objectives</u> ) and manuscripts
Jul. '00:	VLF reports results of studies at annual meetings of the <i>Evolution Society</i> and <i>Society for Conservation Biology</i>
Apr. 15 '01:	VLF submits final report

## **C. Completion Date**

Data collection and analysis will be completed for all species by the end of 1999; final reports and manuscripts summarizing results of the completed projects for each species will be prepared during FY 00.

## **PUBLICATIONS AND REPORTS**

Four major publications will be prepared for publication following completion of the project in FY00; each will report estimates of genetic variability, genetic structure and gene flow for one of the target species. These papers will form the basis for the final report, and will be submitted to international peer-reviewed journals such as *Evolution*, *Molecular Ecology*, or *Auk*, as well as to managers involved with restoration.

## **PROFESSIONAL CONFERENCES**

Interim results from FY98 will be presented as contributed papers by the principal investigators at the annual meetings of the Society for Conservation Biology and the American Ornithological Union in July, 1998 (locations to be announced). Interim and final results in subsequent years will be presented at the Annual Restoration Workshops, as well as at international scientific conferences such as the annual meetings of the Pacific Seabird Group, the American Ornithological Union, the Cooper Ornithological Union, and/or the Society for the Study of Conservation, as well as at the International Ornithological Congress to be held in South Africa in 1998.

## **NORMAL AGENCY MANAGEMENT**

Not applicable.

## **COORDINATION AND INTEGRATION OF RESTORATION EFFORT**

Collection of samples will be coordinated with ongoing studies of seabird feeding ecology in Alaska conducted by the National Biological Service (J.F.P.) and the U.S. Fish and Wildlife Service (Alaska Maritime National Wildlife Refuge). Tissues and skeletons obtained from seabirds will be archived at the American Museum of Natural History (New York), and tissues also will be collected for use in ongoing studies of seabird trophic relationships using stable isotope ratios (K. Hobson, Canadian Wildlife Service, Saskatoon). Samples from carcasses salvaged from the Spill will be obtained from the Burke Museum. This project is made possible by a previous contract for Can\$106,000 awarded to VLF and Dr. Tim Birt by the Environmental Innovations Program of Public Works and Government Services Canada, which enables the development of primers and protocols for 20 nuclear introns and their application to the conservation of marbled murrelets. The present project also is made possible through the donation of tissue samples from murrelets and guillemots by field researchers in Canada and the United States (see **Methods - Population surveys**); these samples are worth an estimated \$10,500.

## **EXPLANATION OF CHANGES IN CONTINUING PROJECTS**

The present proposal differs little from the proposal that was approved in Dec. 1996, but represents a scaled-down version of the original proposal from FY97. Specifically, developmental work included in the original proposal has since been completed under a grant from the Lindbergh Foundation, and fewer loci will be analyzed. These measures reduce the costs both for salaries and for commodities. (Depending on results, more loci may need to be analyzed in FY99.) The amount requested for field collections has also been reduced in the hope that we can obtain more samples through collaborative efforts. (This cost also may have to increase in FY99.) Furthermore, milestones have been altered to accommodate the timing of funding. The present proposal differs from that approved by the Trustee Council in Dec. 1996 in that Queen's University has required the inclusion of a 10% overhead on the total budget

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## PRINCIPAL INVESTIGATORS

Principal investigator - Dr. Vicki Friesen (Assistant Professor of Biology, Queen's University, Kingston, Ontario) completed undergraduate and graduate work in 1992 on the ecology and genetics of seabirds in the North Atlantic. Her doctoral project involved a molecular study of population differentiation and evolution in common and thick-billed murre. She is an author on 23 publications in peer-reviewed scientific journals, including papers on behavioral ecology, genetics and evolution of various vertebrates, primarily seabirds. On-going projects in her research lab include population genetic and phylogenetic studies of murre, murrelets, guillemots, auklets, shags, storm-petrels, ptarmigan and parrots. She will be responsible for supervising the laboratory component of the project, and writing interim and final reports and manuscripts for publication; she will contribute to laboratory work as necessary to keep the project on schedule. Her curriculum vitae is appended.

Principal investigator - Dr. John F. Piatt (Research Biologist GS-13, Alaska Science Center, National Biological Service, Anchorage, AK) obtained a Ph.D. in Marine Biology from Memorial University of Newfoundland in 1987. His dissertation involved seabird-forage fish interactions. Since 1987, he has studied seabirds both at colonies and at sea in the Gulf of Alaska, Aleutian Islands, and Bering and Chukchi seas. He is an author on approximately 50 peer-reviewed scientific publications about seabirds, fish, marine mammals, and effects of oil pollution on marine birds. He will act as the liaison between V.L.F., the EVOS Trustees and other agencies in Alaska, coordinate collection of samples, and assist with writing of reports and manuscripts for publication.

## OTHER KEY PERSONNEL

Technician I (3 years) - Mr. Shane Doran received a B.Sc. [honours] from Queen's University in 1995. His thesis involved the population genetics of blue-eyed shags. Currently he is working part-time for V.L.F. on the population genetics of murrelets. He will be responsible for population screening for common murre (estimated to require ~2.5 years for completion) as well as for management duties necessary for successful completion of the project (e.g. ordering supplies, maintaining radioisotope records, preparing DNA extractions, etc).

Technician II (3 years) - Mr. Jeff Moy received his B.Sc. from Guelph University. His thesis involved a population genetic study of trout. He will be responsible for population screening for pigeon guillemots, marbled murrelets and Kittlitz's murrelets (estimated to require ~2.5 years for completion).

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Table 1. Sites, numbers of samples available, and numbers of samples needed for genetic analyses of murre, murrelets and guillemots.

Site	Avail- able	Needed
<b>COMMON MURRE</b>		
California (Farallon Islands)	30	0
Washington (Clallam)	12	8
N. Vancouver Island	40	0
Southeastern Alaska	0	30
Prince William Sound (Cordova)	23	7
Middleton Island	0	30
Upper Cook Inlet (Kachemak Bay, Chisik I.)	48	0
Lower Cook Inlet (Barren Is.)	27	3
Alaska Peninsula (Semidi, Midun Is.)	18	12
Eastern Aleutians (Aiktak I.)	14	16
Western Aleutians (Attu, Agattu & Buldir Is.)	25	5
Bering Sea (Pribilof, St. Matthew, St. Lawrence Is.)	30	0
Chukchi Sea (Capes Lisburne & Thompson)	33	0
Sea of Okhotsk (Talan I., Magadanskaya)	30	0
Japan (Teuri I.)	0	30
<b>MARBLED MURRELET</b>		
California	12	18
Oregon	12	18
Washington	18	12
British Columbia (Queen Charlotte Is.)	30	0
Southeastern Alaska (Lemesurier I.)	20	10
Prince William Sound (Unakwik Fjord)	15	15
Cook Inlet (Kachemak Bay)	21	9
Kodiak Island	2	18
Mitrofanova Bay	26	4
Shumagin Islands (Koniugi Is., Belofski B., Yakutat P.)	14	16
Central Aleutians (Adak I.)	5	25
Western Aleutians (Attu I.)	9	21

Table 1, cont'd.

Site	Avail- able	Needed
<b>KITTLITZ'S MURRELET</b>		
Prince William Sound	2	*
Kachemak Bay	18	*
Western Aleutians (Attu I.)	5	*
<b>PIGEON GUILLEMOT</b>		
California (Farallon Is.)	20	10
Oregon	25	5
British Columbia (Queen Charlotte Is.)	0	30
Southeast Alaska (Glacier Bay)	0	30
Prince William Sound (Jackpot & Naked Is.)	30	0
Cook Inlet (Kachemak Bay)	9	21
Kodiak Island	0	30
Alaska Peninsula (Semidi and Shumagin Is.)	7	23
Western Aleutians (Attu, Agattu Is.)	0	30
Kuril Is.	0	30
Bering Sea (Pribilof, St. Lawrence Is.)	0	30
Chukchi Sea (Capes Thompson and Lisburne)	0	30

\*Samples will be obtained from Kittlitz's murrelets opportunistically.

NOTE: Every effort will be made to obtain samples non-destructively to minimize the need for collections, e.g. as feathers or blood samples collected during banding, or from museum specimens.

**Victoria Louise Friesen**

**Curriculum Vitae - 11 April 1997**

**Education**

- 7/1992-6/1994      -Postdoctoral Fellow, Ornithology  
                         -Royal Ontario Museum and University of Toronto  
                         -"A phylogenetic analysis of the family Alcidae"
- 5/1987-6/1992      -Doctor of Philosophy, Biopsychology  
                         -Memorial University of Newfoundland, St. John's, Newfoundland  
                         -"Population differentiation and evolution within thick-billed (*Uria lomvia*) and common (*U. aalge*) murres"
- 4/1984-2/1987      -Master of Science, Biopsychology  
                         -Memorial University of Newfoundland, St. John's, Newfoundland  
                         -"Parental energy expenditures and activity budgets of northern gannets (*Sula bassanus*)"
- 9/1980-4/1984      -Bachelor of Science (first class honours), Biology  
                         -University of Prince Edward Island, Charlottetown, Prince Edward Island

**Scholarships and Awards (last five years)**

- 1994                      -NSERC Women's Faculty Award  
                         -Queen's University
- 1992                      -NSERC Postdoctoral Fellowship  
                         -University of Toronto and Royal Ontario Museum

**Manuscripts and Grant Proposals Reviewed For**

-Auk, Canadian Journal of Zoology, Canadian Society for Endangered Birds, Canadian Wildlife Service Special Publications, Condor, Ecology, Evolution, Marine Ecology Progress Series, National Science Foundation, Proceeding of the XXth International Ornithological Congress, Wilson Bulletin

**Major Research Grants and Contracts (last five years)**

- 11/96                      -Advisory Research Council Research Grant  
                         -\$4,000  
                         -"Development of an innovative molecular technique for the conservation of genetic variation in endangered birds"

- 06/96                   -Lindbergh Foundation  
                           -\$10,850 US  
                           -"Development of an innovative molecular technique to aid in balancing technological progress with wildlife management and environmental conservation"
- 11/95                   -Advisory Research Council Research Grant  
                           -\$5,000  
                           -"Equipment for development of a molecular genetic technique for studies in ecology and evolution"
- 08/95-03/97           -Environmental Innovation Program, Public Works and Government Services Canada Contract  
                           -\$106,000  
                           -"Development of an innovative molecular technique for the conservation of genetic variation in endangered birds"
- 05/95                   -Advisory Research Council Research Grant  
                           -\$5,000  
                           -"Development of an innovative genetic technique to aid in studies of ecology and evolution"
- 11/94                   -Advisory Research Council Research Grant  
                           -\$5,000  
                           -"Population differentiation and speciation in guillemots"
- 04/94-03/97           -NSERC Research Grant  
                           -\$35,000  
                           -"Mechanisms of population differentiation and speciation in seabirds"
- 04/94-03/95           -Endangered Species Recovery Fund (World Wildlife Fund)  
                           -\$12,000  
                           -"Conservation genetics of marbled murrelets"

### Refereed Papers

- Kidd, M.G. and V.L. FRIESEN. In revision. Sequence variation in the guillemot (Alcidae: *Cephus*) mitochondrial control region and its nuclear homolog. *Mol. Biol. Evol.*
- FRIESEN, V.L., B.C. Congdon, H.E. Walsh and T.P. Birt. In press. Intron variation in marbled murrelets detected using analyses of single-stranded conformational polymorphisms. *Mol. Ecol.*
- FRIESEN, V.L. and D.J. Anderson. In press. Phylogeny and evolution of the Sulidae: A test of alternative modes of speciation. *Mol. Phyl. Evol.*
- FRIESEN, V.L., W.A. Montevecchi, A.J. Baker, A.J. Gaston, R.T. Barrett and W.S. Davidson. 1996. Population differentiation and evolution in the common guillemot *Uria aalge*. *Mol. Ecol.* 5:793-805.

- FRIESEN, V.L., J.F. Piatt and A.J. Baker. 1996. Evidence from cytochrome *b* sequences and allozymes for a 'new' species of alcid, the long-billed murrelet (*Brachyramphus perdix*). *Condor* 98:681-690.
- FRIESEN, V.L., W.A. Montevecchi, R.T. Barrett and W.S. Davidson. 1996. Molecular evidence for kin groups in the absence of large-scale genetic differentiation in a migratory bird. *Evolution*: 50:924-930.
- FRIESEN, V.L., A.J. Baker and J.F. Piatt. Phylogenetic relationships within the Alcidae (Charadriiformes: Aves) inferred using total molecular evidence. *Mol. Biol. Evol.* 13: 359-367.
- Birt, T.P., V.L. FRIESEN, J.M. Green and W.S. Davidson. Mitochondrial DNA variation in Newfoundland and Norwegian populations of Atlantic capelin, *Mallotus villosus*, detected using two techniques. *Mol. Ecol.* 4: 771-776.
- FRIESEN, V.L., R.T. Barrett, W.A. Montevecchi and W.S. Davidson. 1993. Molecular identification of a backcross between a female common murre/thick-billed murre hybrid and a male common murre. *Can. J. Zool.* 71: 1474-1477.
- FRIESEN, V.L., W.A. Montevecchi and W.S. Davidson. 1993. Cytochrome *b* nucleotide sequence variation among the Atlantic Alcidae. *Hereditas* 119: 245-252..
- BIRT-FRIESEN, V.L., W.A. Montevecchi, A.J. Gaston and W.S. Davidson. 1992. Genetic structure of thick-billed murre (*Uria lomvia*) populations examined using direct sequence analysis of amplified DNA. *Evolution* 67: 267-272.
- Birt, T.P., V.L. FRIESEN, J.M. Green, W.A. Montevecchi and W.S. Davidson. 1992. Cytochrome *b* variation in parrots. *Hereditas* 117: 67-72.
- Montevecchi, W.A., V.L. BIRT-FRIESEN and D.K. Cairns. 1992. Reproductive energetics and prey harvest of Leach's storm-petrels in the northwest Atlantic. *Ecology* 73: 823-832.
- Cairns, D.K., W.A. Montevecchi, V.L. BIRT-FRIESEN and S.A. Macko. 1990. Energy expenditures, activity budgets, and prey harvest of breeding common murres. *St. Av. Biol.* 14: 84-92.
- BIRT-FRIESEN, V.L., W.A. Montevecchi, D.K. Cairns and S.A. Macko. 1989. Activity-specific metabolic rates of free-living northern gannets and other seabirds. *Ecology* 70: 357-367.
- Davidson, W.S., S.E. Bartlett, T.P. Birt, V.L. BIRT and J.M. Green. 1989. Identification and purification of serum albumin from rainbow trout (*Salmo gairdneri*). *Comp. Biochem. Physiol.* 93B: 5-9.
- Davidson, W.S., V.L. BIRT, T.P. Birt and J.M. Green. 1988. Palmitate-binding, serum albumin-like proteins in salmonids. *FEBS Letters* 233: 299-302.
- Montevecchi, W.A., V.L. BIRT and D.K. Cairns. 1988. Dietary changes of seabirds associated with local fisheries failures. *Biol. Oceanogr.* 5: 153-161.
- Montevecchi, W.A., D.K. Cairns and V.L. BIRT. 1988. Migration of post-smolt Atlantic salmon, *Salmo salar*, off northeastern Newfoundland, as inferred by tag recoveries in a seabird colony. *Can. J. Fish. Aquat. Sci.* 45: 568-571.
- BIRT, V.L., T.P. Birt, D. Goulet, D.K. Cairns and W.A. Montevecchi. 1987. Ashmole's halo: direct evidence for prey depletion by a seabird. *Mar. Ecol. Prog. Ser.* 40: 205-208.
- BIRT, V.L. and D.K. Cairns. 1987. Kleptoparasitic interaction of arctic skuas *Stercorarius parasiticus* and black guillemots *Cephus grylle* in northeastern Hudson Bay, Canada. *Ibis* 29: 190-196.
- Cairns, D.K., K.A. Bredin and V.L. BIRT. 1987. A tunnel for hidden access to blinds at high latitude seabird colonies. *J. Field Ornithol.* 58: 69-72.



Cairns, D.K., K.A. Bredin, V.L. BIRT and W.A. Montevecchi. 1987. Electronic activity recorders for aquatic wildlife. *J. Wildl. Manag.* 51: 395-399.

### **Papers Submitted or In Preparation**

- FRIESEN, V.L., J.F. Piatt and A.J. Baker. Molecular insights into the evolution of the Alcidae. In prep. for *Mol. Phyl. Evol.*
- FRIESEN, V.L., D.K. Cairns, W.A. Montevecchi and S.A. Macko. Parental time and energy budgets of northern gannets (*Sula bassana*). In prep. for *Physiol. Zool.*
- Kidd, M.G. and V.L. FRIESEN. Patterns of control region variation in populations of *Cephus guillemots*: testing microevolutionary hypotheses. In prep. for *Evolution*.
- Montevecchi, W.A., R.E. Ricklefs and V.L. FRIESEN. Organic composition and condition of nestling and fledgling northern gannets. In prep. for *Can. J. Zool.*

### **Scientific Presentations**

- Kidd, M.G. and V.L. FRIESEN. 1997. Patterns of control region variation in populations of *Cephus guillemots*: testing microevolutionary hypotheses. *24th Meet. Pacif. Seabird Gr.* Oral presentation.
- Walsh, H.E. and V.L. FRIESEN. 1997. Phylogenetic relationships among the auklets. *24th Meet. Pacif. Seabird Gr.* Poster presentation.
- FRIESEN, V.L. 1996. Population genetics and the spatial scale of conservation of colonial waterbirds. *20th Stated Meeting of the Colonial Waterbird Society*. Invited presentation.
- FRIESEN, V.L. 1995. Molecular evidence for kin groups within a colony of thick-billed murres. *Proc. Jt. Conf. Colon. Waterbird Soc. Pac. Seabird Gr.*
- FRIESEN, V.L., H. Jones and D.J. Anderson. 1995. Phylogeny and evolution of the Sulidae. *Proc. Jt. Conf. Colon. Waterbird Soc. Pac. Seabird Gr.*
- Kidd, M.G. and V.L. FRIESEN. 1995. A guillemot intraspecific phylogeny: inferring population history from patterns of geographic variation in *Cephus* mitochondrial control regions. *Proc. Jt. Conf. Colon. Waterbird Soc. Pac. Seabird Gr.*
- FRIESEN, V.L., A.J. Baker and J.F. Piatt. 1995. Conservation genetics of marbled murrelets. *22nd Meet. Pacif. Seabird Gr.* Oral presentation.
- FRIESEN, V.L., A.J. Baker and J.F. Piatt. 1994. Molecular insights into modes of speciation in the Alcidae. *XXI Internal. Ornithol. Congr.* Symposium presentation.
- FRIESEN, V.L., A.J. Baker and J.F. Piatt. 1994. Molecular evidence for a 'new' species of alcid, the long-billed murrelet. *XXI Internal. Ornithol. Congr.* Poster presentation.
- FRIESEN, V.L., A. J. Baker and J.F. Piatt. 1994. A molecular investigation of evolutionary relationships within the Alcidae. *21st Ann. Meet. Pacif. Seabird Gr.* Opening presentation.
- FRIESEN, V.L., W.A. Montevecchi and W.S. Davidson. 1994. Population genetics and conservation of rare seabirds. *21st Ann. Meet. Pacif. Seabird Gr.* Invited presentation.
- BIRT-FRIESEN, V.L., A.J. Baker and J.F. Piatt. 1993. A phylogenetic analysis of the Alcidae. *111th Meet. Am. Ornithol. Union.* Oral presentation.
- FRIESEN, V.L., W.A. Montevecchi, A.J. Baker and W.S. Davidson. 1993. Population differentiation and phylogeography in two species of murres (genus *Uria*). *32nd Ann. Meet. Can. Soc. Zool.*

Oral presentation.

- FRIESEN, V.L., W.A. Montevecchi and W.S. Davidson. 1993. Genetic substructuring within a colony of thick-billed murres. *1st Int. Meet. Wilson Ornithol. Soc. Can. Soc. Ornithol.* Oral presentation.
- FRIESEN, V.L. and A.J. Baker. 1992. Utility of mitochondrial DNA sequencing in conservation biology. *54th Midwest Fish Wildl. Confer.* Invited presentation.
- BIRT-FRIESEN, V.L., W.A. Montevecchi and W.S. Davidson. 1990. Multi-level analyses of genetic affinities within the genus *Sula*. *XX Internatl. Ornithol. Congr.* Oral presentation.
- BIRT-FRIESEN, V.L., W.A. Montevecchi, A.J. Gaston and W.S. Davidson. 1990. Inter-colony genetic comparison of thick-billed murres (*Uria lomvia*) using the polymerase chain reaction. *Col. Waterbird Soc. Ann. Meet.* Invited presentation.
- Montevecchi, W.A., V.L. BIRT-FRIESEN and D.K. Cairns. 1990. Avian energetics and bioindication in the northwestern Atlantic. *Col. Waterbird Soc. Ann. Meet.* Invited presentation.
- BIRT-FRIESEN, V.L., W.A. Montevecchi and W.S. Davidson. 1989. Development of molecular genetic techniques to probe the population structure of thick-billed murres in the North Atlantic. *Pop. Biol. Conserv. Mar. Birds Workshop.* Oral presentation.
- BIRT, V.L., W.A. Montevecchi & W.S. Davidson. Aug. 1988. Novel DNA probes for seabird population genetics studies. *XIX Internatl. Congr. Genet. Genome* 30, Suppl. 1: 371. Poster.
- BIRT, V.L., T.P. Birt, D. Goulet, D.K. Cairns & W.A. Montevecchi. March 1986. Ashmole's halo: direct evidence for prey depletion by a seabird. *Pop. Biol. Conserv. Mar. Birds, Proc., Pac. Seabird Gr. Bull.* 13: 100. Oral presentation.
- BIRT, V.L., D.K. Cairns, S.A. Macko & W.A. Montevecchi. March 1986. Energetics of free-ranging northern gannets during the breeding season. *Pop. Biol. Conserv. Mar. Birds, Proc., Pac. Seabird Gr. Bull.* 13:102. Oral presentation.
- FRIESEN, V.L., D.K. Cairns, S.A. Macko & W.A. Montevecchi. June 1986. Parental energy expenditures of free-ranging northern gannets and common murres. *XIX Internatl. Ornithol. Congr.* Poster.

# 1998 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1997 - September 30, 1998

Budget Category:	Authorized FY 1997	Proposed FY 1998						
Personnel	\$0.0	\$5,946.0						
Travel	\$0.0	\$5,500.0						
Contractual	\$56,000.0	\$70,500.0						
Commodities	\$0.0	\$500.0						
Equipment	\$0.0	\$0.0	LONG RANGE FUNDING REQUIREMENTS					
Subtotal	\$56,000.0	\$82,446.0		Estimated FY 1999	Estimated FY 2000	Estimated FY 2001	Estimated FY 2002	
General Administration	\$0.0	\$2,314.4						
Project Total	\$56,000.0	\$84,760.4		\$86.2	\$13.8	\$0.0		
Full-time Equivalents (FTE)	0.0	0.3						
Dollar amounts are shown in thousands of dollars.								
Other Resources								
Comments:								
2.6% of the budget is for conference travel.								
This budget represents an increase over the predicted budget for FY98 due to inclusion of a 10% overhead for Queen's University and an administrative charge of \$2313 for DOI.								

1998

Prepared: 14 Apr 1997

Project Number: 98-169  
 Project Title: A genetic study to aid in restoration of murre, guillemots and murrelets to the Gulf of Alaska  
 Agency: Queen's University (VLF) and DOI (JFP)

FORM 3A  
 TRUSTEE  
 AGENCY  
 SUMMARY

4/14/97

**October 1, 1997 - September 30, 1998**

1998

Project Number: 98-169  
Project Title: A genetic study to aid in restoration of murrelets, guillemots and murrelets to the Gulf of Alaska  
Agency: Queen's University (VLF) and DOI (JFP)

FORM 3B  
Personnel  
& Travel  
DETAIL

October 1, 1997 - September 30, 1998

1998

Agency: Queen's University (VLF) and DOI (JFP)

**FORM 3B**  
**Contractual &**  
**Commodities**  
**DETAIL**

**October 1, 1997 - September 30, 1998**

1998

# FORM 3B Equipment DETAIL

**1997 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET**  
October 1, 1996 - September 30, 1997

Budget Category:	Authorized FFY 1996	Proposed FFY 1997						
Personnel	\$36,000.0	\$48,000.0						
Travel	\$1,100.0	\$2,200.0						
Contractual								
Commodities	\$10,200.0	\$13,600.0						
Equipment	\$8,000.0		<b>LONG RANGE FUNDING REQUIREMENTS</b>					
Subtotal	\$55,300.0	\$63,800.0	Estimated FFY 1999	Estimated FFY 2000	Estimated FFY 2001	Estimated FFY 2002	Estimated FFY 2003	
Indirect	\$200.0	\$6,900.0						
Project Total	\$55,500.0	\$70,500.0	\$86.2	\$13.8				
Full-time Equivalents (FTE)	1.5	2.0						
Other Resources			Dollar amounts are shown in thousands of dollars.					

All estimates assume a U.S./Canadian exchange rate of 0.74.

Indirect costs include costs for Xeroxing, telephone calls, shipping samples, permits, etc (approx. \$300) and 10% overhead for Queen's University.

**1998**

Prepared: 10 Apr. 1997

Project Number: 98-169  
Project Title: A genetic study to aid in restoration of murre, guillemots and murrelets to the Gulf of Alaska  
Name: Queen's University (V.L. Friesen) and DOI (J.F. Piatt)

**FORM 4A  
Non-Trustee  
SUMMARY**

4/14/97

October 1, 1996 - September 30, 1997

1998

FORM 4B  
Personnel  
& Travel  
DETAIL



**1997 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET**

October 1, 1996 - September 30, 1997

<b>Contractual Costs:</b>		<b>Proposed FFY 1997</b>
<b>Description</b>		
<b>Contractual Total</b>		<b>\$0.0</b>
<b>Commodities Costs:</b>		<b>Proposed FFY 1997</b>
<b>Description</b>		
CONSUMABLE AND DISPOSIBLES*		
Technician I		6,800.0
Technician II		6,800.0
<p>*Each technician will screen DNA from approximately 225 samples for each of 20 genes. Each sample costs \$0.90/gene for amplifications with incorporation of 33P-dATP, and \$0.55/sample for SSCP or microsatellite gels. Thus each technician will require ~\$6525 for reagents for population screening. Approximately \$250/yr will be needed for gloves etc, for an annual cost of \$6775 each.</p>		
<b>Commodities Total</b>		<b>\$13,600.0</b>

**1998**

Project Number: 98-169  
 Project Title: A genetic study to aid in restoration of murre, guillemots and murrelets to the Gulf of Alaska  
 Name: Queen's University (V.L. Friesen) and DOI (J.F. Piatt)

**FORM 4B  
 Contractual &  
 Commodities  
 DETAIL**

## 1997 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

**October 1, 1996 - September 30, 1997**

New Equipment Purchases:		Number of Units	Unit Price	Proposed FFY 1997
Description				
				0.0
				0.0
				0.0
				0.0
				0.0
	Note: This project is made possible by the existence of equipment valued in excess of \$50,000 within VLF's DNA research laboratory at Queen's University			0.0
	The requested equipment is required to handle additional demand on apparatus associated with the proposed project			0.0
				0.0
				0.0
				0.0
				0.0
Those purchases associated with replacement equipment should be indicated by placement of an R.			<b>New Equipment Total</b>	<b>\$0.0</b>
Existing Equipment Usage:			Number of Units	
Description				

# 1998

Project Number: 98-169

Project Title: A genetic study to aid in restoration of murre, guillemots and murrelets to the Gulf of Alaska

Name: Queen's University (V.L. Friesen) and DOI (J.F. Piatt)

FORM 4B  
Equipment  
DETAIL



## Isotope Ratio Studies of Marine Mammals in Prince William Sound

Project Number: 98170

Restoration Category: Research

Proposer: University of Alaska Fairbanks

Lead Trustee Agency: ADFG

Cooperating Agencies: None

Alaska SeaLife Center:

Duration: 3rd year, 3-year project

Cost FY 98: \$111,530

Geographic Area: Prince William Sound/Gulf of Alaska

Injured Resource/Service: Harbor seals, nearshore ecosystem species, seabirds

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APR 15 1997

EXXON VALDEZ OIL SPILL  
TRUSTEE COUNCIL

### ABSTRACT

This project uses natural stable isotope ratios to assess trophic structure and food webs in Prince William Sound and contributes to studies by ADFG personnel to determine the reasons for the decline of harbor seal populations. Through a mix of captive animal studies and a comparison of isotope ratios in prey species and archived and current marine mammal tissues, insight into environmental changes causing the decline may be possible. Preliminary data point strongly toward a major decline in the carrying capacity of the northern Pacific Ocean in the past two decades. This decline is evident in the abundance and distribution of marine biota and is reflected in the carbon isotope ratios of marine mammals of the region.

## INTRODUCTION

Through FY 98 this study will conclude an assessment, via isotope ratio techniques, of trophic energetics supporting marine mammals in the Prince William Sound (PWS) region. The results of the study to date imply that over the past two decades the carrying capacity of the western Gulf of Alaska (including Prince William Sound) and the Bering Sea has declined by 35–40 percent. This major loss of primary production is implied from an observed decline in carbon isotope ratios, which have been shown by others to be in direct response to average phytoplankton growth rates. This loss of carrying capacity may be a major reason for the observed continuing declines in the marine mammal populations of the region, especially harbor seals and Steller sea lions. There remain many uncertainties — the couplings of decreased primary productivity up the food chain are complex and some species do not appear affected. The underlying causes probably arise from changes in the physical and chemical environment and may be cyclical or in response to a longer-term climatic change.

Our work over the past three years has provided a rapidly growing data base which is now being synthesized to define marine mammal ecosystem interactions in Prince William Sound and surrounding waters. Stable isotope tracers have provided a means of following advection into the sound from offshore and of identifying geographic separations in the food webs supporting some of the injured species, primarily sea otters, harbor seals and sea birds. Our methods and procedures continue as proposed in FY 97 and are included in this proposal for convenience of the reader. An accompanying Annual Progress Report details the accomplishments to date and the preliminary findings.

Over the past two decades, isotope ratio analysis has emerged as a powerful tool in ecosystem research on the process scale and as a validation technique for large-scale ecosystem models (Michener and Schell, 1994). In applications relevant to this study, Schell et al. (1989a) described a geographic gradient in isotope ratios in biota across the Alaskan Beaufort Sea and the Bering-Chukchi seas and showed that this gradient could be applied to describing bowhead whale natural history. The isotopic gradient arises from the primary producers in the ecosystem and is passed up food chains to label consumers up to the top predators. Saupe et al. (1989) describes the parallel shifts in  $\delta^{13}\text{C}$  in euphausiids and copepods across this region, and Schell et al. (1989b, 1992) discussed the effects of the gradient in forming oscillations in isotope ratios in whale baleen. Hobson and Welch (1992) used isotope ratios to describe the trophic relationships of birds and mammals to the available prey species in the Canadian Arctic. Further extension to benthos by Dunton et al. (1991) and to fishes (Vinette, 1992) has confirmed that the isotopic trends are evident across the entire food web.

In contrast to the primarily geographic control on carbon isotope ratios, nitrogen isotope ratios are influenced by trophic level. Vinette (1992) has shown that the  $\delta^{15}\text{N}$  of euphausiids and copepods in the continental shelf regions of the Bering, Chukchi and Beaufort seas are statistically indistinguishable. Within a given region, when pelagic and benthic species of known feeding habits are compared, a predictable enrichment in  $^{15}\text{N}$  of approximately 3.3% per trophic level increase occurs. By assembling the trophic spectrum of species within an ecosystem it is possible to ascribe status within the ecosystem. Hobson and Welch (1992) were able to use  $\delta^{15}\text{N}$

values in the Barrow Strait–Lancaster Sound region to identify the role of arctic cod (*Boreogadus saida*) and other prey species relative to top consumers. Higher trophic levels showed little change in  $\delta^{13}\text{C}$  but varied by an average of 3.8‰ between levels. Recently, Sease et al. (1993) showed preliminary data that confirmed that sea lions occupy a high trophic status in North Pacific food webs and reflect a geographic gradient between Prince William Sound and the Washington coast.

Our recent work in the Bering Sea and in the Prince William Sound and Gulf of Alaska has shown that pronounced geographic gradients in isotope ratios exist in  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  across the shelf break. This discovery and the acquisition of sufficient data to enable contour mapping of isotopic regimes are now being used to interpret isotope ratio patterns found in Bering Sea sea lions and Gulf of Alaska and Prince William Sound harbor seals. This is requiring the development of statistical approaches and will be a focus of the FY 98 work.

The trophic changes as indicated by  $\delta^{15}\text{N}$  values in seal tissues that may have occurred in the populations of harbor seals are also proving interesting. Samples of tissues from pre-decline seals have been compared with post-decline seals and very little difference is apparent in marked contrast to carbon isotope ratios. Only a limited number of seals have been compared to date—this work is continuing.

Trophic energetics of individual seals can be assessed on a seasonal basis by changes in isotope ratios along the lengths of the vibrissae. Whereas an individual animal may show little temporal change in trophic status as indicated by  $\delta^{15}\text{N}$  values or regional feeding as evidenced by carbon isotope ratios along the length of vibrissae, there have been remarkable differences from one animal to the next in the individuals examined to date.

#### *Preliminary Findings*

Funding for this work resumed in February 1995 (no interim funds from October 1994 were allocated) and we have now accumulated a large data base, presented the findings at professional meetings and submitted a manuscript detailing isotope ratio gradients in the western Gulf of Alaska and the Bering Sea to the *Marine Ecology Progress Series*. Another manuscript, detailing temporal changes in isotope ratios in the environment and the implications for ecosystem carrying capacity, is in preparation. We have completed the first major suite of prey species isotope ratio analyses and collected a wide spectrum of marine mammal samples from Native harvests and through strandings and collections being conducted by ADFG. These samples have been analyzed and are now being synthesized in context with the lower trophic data collected by Tom Kline of the Prince William Sound Science Center. This work is also part of Amy Hiron's Ph. D. dissertation.

A major requirement in interpreting the isotope ratios along the vibrissae of seals as temporal markers is that the growth rates must be known. We are currently conducting calibration experiments in cooperation with the chief veterinarian of the Mystic Marineline Aquarium (MMA), Mystic, Connecticut. The personnel of MMA and the Principal Investigator have agreed upon protocols for undertaking experiments on captive sea lions and harbor seals aimed at the determination of whisker growth rates, diet fractionation factors arising from differing types of prey species, and seasonal cycles in isotope ratios caused by physiological effects. This study has

provided insight into whisker growth rates for harbor seals and Steller sea lions and is still underway. Preliminary data indicate that the vibrissae of Steller sea lions grow at a relatively constant rate over the year whereas harbor seal vibrissae have a strong seasonal response with rapid growth in the spring and summer and slow rates in fall and winter. Experiments are currently underway to determine the effectiveness of oral dosing of isotopically labeled food in contrast to intravenous administration of labeled amino acids. An effective oral dosing and incorporation of the isotopic label would allow more frequent marking. Current procedure involves capture, restraining and holding of the harbor seals, which is complex and labor intensive.

In addition, as part of a synergistic study on Bering Sea marine mammals, we have also conducted the first set of measurements of whisker growth rates on juvenile sea lions being raised at the Vancouver Aquarium in cooperation with Dr. Andrew Trites of the University of British Columbia. These animals are to be moved to the Alaska SeaLife Center, and may be used for further natural history studies in the future.

## **NEED FOR THE PROJECT**

### **A. Statement of Problem**

Harbor seals were undergoing an unexplained decline in numbers before the oil spill and the decline was further accelerated by the disaster. Since that time the population has not recovered and is still at a low level, although now perhaps finally stabilized. No definitive cause and effect relationship has been found for the decline or failure to recover. This project uses stable isotope ratios as natural tracers to test hypotheses regarding shifts in diet or trophic status in the previous decade(s) as underlying reasons for the decline.

A second requirement is to provide isotope ratio analyses for this study and other restoration projects needing isotope abundance information. Based upon samples received during 1996–97, we anticipate a total of approximately 5,000–8,000 samples for isotope ratio analysis in the coming year, a slightly lower number than in the past.

### **B. Rationale/Link to Restoration**

Carbon isotope ratios serve as conservative tracers of energy supply between trophic levels (phytoplankton to zooplankton to fishes to top consumers). Seals, cetaceans, birds, etc., acquire the isotope ratios in proportion to the amount of food derived from each differing source. This, in turn, is reflected in the composition of body tissues and in keratinous tissues (claws, feathers, baleen, whiskers) as a temporal record when multiple sources of food are consumed over time and space. This allows the discerning of important habitats and food resources in animals that seasonally migrate or undergo periods of hyper- and hypotrophy. Carbon isotope ratios have also been closely tied to rates of cell growth by phytoplankton and thus provide an indirect method of assessing seasonal productivity via primary consumers in situations where multi-year samples are available.

Nitrogen isotope ratios reflect both the food sources and the trophic status of that animal. As nitrogen in food is consumed and assimilated by a consumer, the heavy isotope is enriched by approximately 3‰ with accompanying loss of the lighter isotope through excretion. The enrichment occurs with each trophic step and allows the construction of conceptual models and food webs and the assignment of relative trophic status to species for which dietary data are sparse. The data obtained from these measurements are unique in that they trace materials actually assimilated and can be used for more accurate ecosystem modeling.

The availability of macrozooplankton forage for salmon, herring, and their predators varies in space and time because of changes in the physico-chemical processes in Prince William Sound. In the SEA context, the latter is known as the Lake/River processes (SEA hypothesis number 2). When macrozooplankton are not available, consumers are forced to switch prey, the Predator/Prey Relationships (SEA hypothesis number 3) shift in time and space. These shifts represent fundamental changes in the way the PWS ecosystem produces commercial species (i.e. herring and salmon). A better understanding, particularly a quantitative understanding, is a prerequisite to determining protocols for restoration and recovery of these species.

It can be postulated that the natural stable isotope abundances of PWS biota will shift because of changes in trophic level, food web structure, and primary productivity in the context of the SEA hypotheses, thus providing an independent tool to verify, quantify and model ecosystem processes. The tracer nature of the approach will enable the integration of ecosystem components. It will enable us to monitor both "top-down" (predation) and "bottom-up" (food supply) controls on herring and salmon production.

### **C. Location**

The research effort will be conducted in Prince William Sound with contrasting data obtained from samples from the Kodiak Island area and in the coastal Gulf of Alaska near Cordova. Comparative work involving prey items and marine mammals from outside Prince William Sound will be made on cruises of opportunity in the Gulf of Alaska. Calibration experiments on whisker growth rates and diet/stable isotope ratio changes will be undertaken using captive harbor seals at research facilities at the Mystic Marinelife Aquarium, Mystic, Connecticut. The benefits of this project will be realized throughout PWS and will be applicable to other areas of the state.

## **COMMUNITY INVOLVEMENT AND TRADITIONAL ECOLOGICAL KNOWLEDGE**

Community involvement in this project is essential in that a large fraction of the samples will be provided via Native harvests of marine mammals. Kate Wynne of the School of Fisheries and Ocean Sciences, UAF, has collected seal whiskers and tissues for this study in the past and we anticipate this assistance will continue. These samples, which have been archived at the University of Alaska Museum, are now being accessed for tissues as needed.



## PROJECT DESIGN

### A. Objectives

The objectives to be completed during the period of this proposal are essentially the same as in our FY 97 proposal and include:

1. Collect samples of harbor seal vibrissae through continued cooperative work with ADFG and Native hunter programs in Prince William Sound.
2. Collect samples of harbor seal prey species including forage fishes, salmon and herring in the vicinity of major haul-outs and high population densities. Samples of seal tissues will be collected from Native hunters. These samples will be obtained with assistance from ADFG personnel monitoring harvests, and the forage fish samples will be collected by T. Kline.
3. Perform stable isotope ratio analyses on tissues and organisms collected during the sampling program. Through the use of **carbon** isotope data on taxa collected over geographical regions, the presence/absence of **isotopic gradients** useful in sorting out habitat dependencies will be determined. This work is complete for the western Gulf of Alaska and is currently underway for the eastern section.
4. Assist other research programs in the Prince William Sound ecosystem study by conducting stable isotope ratio analyses on samples provided and aid the interpretation of results. This effort will require approximately 20% of the analytical and research effort.
5. Through the use of **nitrogen** isotope ratios in collected taxa, assign **trophic status** to species in each region. Compare trophic status with predictive models based on conceptual food webs.
6. Determine temporal changes in harbor seal trophic status and food dependencies by comparing isotope ratios along the lengths of vibrissae with prey availability and their isotope ratios. Establish the relationships between whisker growth rate and temporal changes and the fractionation factors between the  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  values of diet and consumer through the use of captive animals being fed known diets.
7. Compare the isotope-ratio derived food web models to predictions by the Lake/River hypothesis and others being tested by the SEA project as an independent means of validation.

### B. Methods

The primary work will be divided into the sampling program and the subsequent analytical and synthesis tasks. Sampling of tissues for stable isotope analysis has been described for both bulk tissues (muscle, blubber) and temporally variable tissues (whiskers, claws, etc.) (Schell et al. 1989; Michener and Schell, 1994).

1. Analytical — Vibrissae from seals, either from Prince William Sound or captive animals, are noted for facial location. A whisker is then segmented at 2.5 mm intervals with a razor and subsamples are placed in vials for later grinding and mass spectrometry. Subsamples are dried and powdered for homogeneity and isotope ratios of carbon and nitrogen are determined with a Europa 20/20 mass spectrometer system. The sample is flash combusted at high temperature and nitrogen and carbon dioxide gases are separated and purified by gas chromatography. These are subsequently injected into the mass spectrometer by capillary and the isotope ratios are determined. The analytical replicability for the entire sampling process is better than  $\pm 0.05\%$  for both  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$ .
2. Sampling — The acquisition of samples for isotope analysis will be conducted through several channels. Forage fish, pollock and other commercial species will be obtained through cooperative programs with the National Marine Fisheries Service, ADFG, and from the Prince William Sound Science Center. As part of the cooperative effort with Dr. Kline, samples will be recorded and the analyses run on a coordinated suite of specimens collected over the geographic regions of the PWS and throughout the seasons. This will allow "within taxa" comparisons to determination shifts in trophic levels and discrimination of the effects of geographic shifts of isotope ratios in primary producers.

Samples of marine mammals, birds, etc., have been and will be obtained from archived materials, strandings, Native harvests and, in some cases, collection in the field. This effort will be closely coordinated with the U.S. Fish and Wildlife Service, ADFG, and the EVOS-sponsored efforts having field programs. Our experience has already produced a wide variety of samples and there is reason to anticipate that 1997-98 will be even more productive. The small amount of sample required for isotopic analyses means that little effort for preservation or transport is required.

The application of isotope ratio work to marine mammals is relatively new and the technique is still in the process of calibration. We have been offered the opportunity to conduct captive animal experiments at the Mystic Marinelife Aquarium in Connecticut using Steller sea lions and harbor seals. Seal vibrissae have been marked through the intravenous addition of  $^{15}\text{N}$  and  $^{13}\text{C}$  labeled amino acids, and growth rates have been measured over the seasonal cycles to determine if physiological effects are translated into differing isotope ratios. This work is continuing but will comprise only a limited amount of the total effort. It is, however, essential given this relatively new field of application. Future studies are planned for the Alaska SeaLife Center at Seward in 1998.

Cooperating investigators will supply the PI with tissue samples for analysis in the form of dry powdered material to expedite handling and analysis. If samples must be prepared by the personnel in the PI's laboratory, a charge for preparation will be made to the investigator or a reduced number of samples will be run depending on the difficulties involved. Similarly, glass fiber filtered samples will be charged at an increased rate because of the accelerated destruction of the combustion furnace tubes from the melted glass particles. Since almost all sample materials are dried tissues, no significant problems are anticipated in this respect.

3. Synthesis of data — The plots of isotope ratios of carbon and nitrogen along the lengths of vibrissae from harbor seals are known to show oscillations in isotope ratios in response to dietary changes over the season (Schell, 1993–95 data). As new data with supporting natural history information are acquired, the values at specific intervals will be compared with potential prey for likely matches. These will be compared with observational data and known feeding habits. From this information sampling can be constrained to the most probable food sources and further directed analyses performed to confirm or deny conceptual food web structure. In cooperation with ADFG personnel, stable isotope data will also be compared with fatty acid compositions in seal blubber to determine if other proxies for dietary components can be established.

Additional synthesis efforts will be made in conjunction with modeling projects associated with the SEA program. The data we acquire is very valuable in that it is an independent means of validating food web and energy flow models to top consumers. If isotopic data are in conflict with that projected from the model calculations, it is usually the model that is in error. Although a complex ecosystem such as Prince William Sound, with robust interactions between land and sea can give rise to varied isotopic abundances in the biotic components, the strong integrating effects that occur in building the “whole body” are very amenable to stable isotope tracers.

### **C. Cooperating Agencies, Contracts, and Other Agency Assistance**

None

## **SCHEDULE**

### **A. Measurable Project Tasks for FY 98 (October 1, 1997 – September 30, 1998)**

October - February:	Preparation and analysis of isotope ratio samples collected in 1994-1996
December:	Collection of vibrissae from isotopically labeled seals and sea lions at Mystic, Connecticut
15 February - 31 March:	Synthesis and coordination for sampling in 1997. Annual report on FY 97 (and prior) work
April - August:	Preparation of draft manuscripts and final report, captive animal experiments
August - September:	Data synthesis, identification of gaps. Manuscript preparation, Final Report submission

### **B. Project Milestones and Endpoints**

The milestones in this project are a blend of definitive goals and a continuing research process that will extend to the end of the funding period. Specific goals follow:

*Captive animal studies* of vibrissae growth rates and dietary effects on stable isotope ratios — Now underway, completion anticipated in spring 1998. This is later than originally proposed because of erroneous assumptions about growth rates that now need to be corrected via altered experimental protocols. Harbor seals grow vibrissae seasonally, whereas those of sea lions grow at a relatively continuous rate.

*Field collections of prey species over the geographic region, collections of whiskers and tissues from harbor seals* — Currently underway. Will continue through FY 98 but will be more directed toward the end of the study as we fill data gaps.

*Stable isotope analyses* — The laboratory work associated with the preparation of samples and isotope ratio analyses will continue throughout the duration of this project but will become more focused as the end approaches. The major collection and data base construction will occur during FY 98.

*Modeling and synthesis of results* — This will occur over the entire project in an iterative process with the emphasis building in FY 98 and continuing until the conclusion of the project.

*Assistance to other investigators* — This aspect is now underway and will continue throughout the project. It is anticipated that the maximum interaction occurred during FY 97 but demand for sample analysis and interpretation is continuing. Synthesis and interpretation of isotope ratio data will be ongoing.

#### Project milestones and reporting periods:

October 1997 - February 1998:	Analysis of 1997 field season samples. Preparation of journal manuscripts
March 1998 - April 1998:	Continue analyses
April 1998 - December 1998:	Draft final report, continue analytical work
January 1999 - March 1999:	Final report, synthesis meetings, manuscript preparation

#### C. Completion Date

The sampling and analytical aspects of this project are anticipated to be completed in 1998. The service aspects of the mass spectrometry for isotope ratios may continue beyond that date if demand warrants.

#### PUBLICATIONS AND REPORTS

Results of this project will be made available via the following:

*Annual Reports:* These reports will detail progress, preliminary findings and notable achievements. These are anticipated for the ends of FY 96 and FY 97.

*Final Report:* A Final Report will be provided. Technical results in these reports will be shared with EVOS collaborators. Preliminary exchanges of findings will be conducted with EVOS investigators and the scientific community via professional meetings and informal communications. The PI will provide expertise in interpretation of isotope results to other projects for which the isotope techniques are only a minor portion of the scientific effort. The final reports of the PI will assist others in that they will provide independent means for validation of trophic models and energy flow descriptions of the Prince William Sound ecosystem.

*Peer-reviewed publications:* Over the course of this study peer-reviewed publications will be generated for the open literature based on the scientific findings. These publications will be generated by the PI and graduate students as first author publications where the primary focus is on the findings produced by the isotopic techniques or as second author publications when the isotope work is a minor part of the scientific results.

*Papers at scientific society meetings:* We request support for travel to appropriate scientific meetings for dissemination of results and interaction with colleagues. It is anticipated that the Society for Marine Mammalogy or the American Society for Limnology and Oceanography meetings will be attended by the PI and graduate student Amy Hirons. The Marine Mammal Meeting in January 1998 is in Monaco and the added costs over a domestic meeting will be obtained from other sources.

*Public Lectures:* Interaction with the public will arise through formal and informal presentations of results. Synthesis meetings designed to explain the findings of ecosystem studies will be presented at meetings coordinated by the EVOS program and open to the public. Informal presentation of results will occur through interaction with interested members of the public, press and scientific community. Classroom instruction will also involve integration of findings into the presentation of educational material.

## **PROFESSIONAL CONFERENCES**

The results of this project were presented at the joint American Society of Limnology and Oceanography/American Geophysical Union meeting in Santa Fe, January 1997. As noted above, the biennial meeting of the Society for Marine Mammalogy is to be held next in Monaco and the costs for this travel will be shared between EVOS and other sources.

## **COORDINATION AND INTEGRATION OF RESTORATION EFFORT**

1. **Resources and Services** — This study focuses on harbor seals, sea birds and the cetaceans of Prince William Sound. Although the major effort is concerned with harbor seals, other marine mammal tissues will be collected in cooperation with those agencies handling or collecting those species. The principal cooperating agency personnel are Kathy Frost and co-workers of ADFG, with whom a wide variety of sampling efforts have already been

undertaken and are continuing in 1995. Dr. Michael Castellini and Brian Fadely have also provided invaluable help by accessing whiskers from seals in their tagging program.

2. Relations to Other Damage Assessment Work — This study is closely coordinated with the modeling efforts and the pelagic food web studies being undertaken by Prince William Sound Science Center personnel. Dr. Kline is responsible for most pelagic collections of food base organisms and is sharing these data to help construct the food web models. Dr. Schell is responsible for the marine mammal aspects and will collect additional forage species as required by his project (for example, samples of herring, capelin, sand lance, etc., in regions of high marine mammal density or active feeding). Stable isotope data provide an excellent means for validating models and testing food web linkages. This aspect of the work will be cooperative with many components of the SEA project.

We are very fortunate to be simultaneously involved in an isotope study on marine mammals in the Bering Sea. This project, which has been supported by the North Pacific Universities Consortium in the past and is currently funded by the Coastal Marine Institute, will provide a valuable amount of complementary data and assist in gathering insight as to the mechanisms involved in the marine mammal population declines.

## **EXPLANATION OF CHANGES IN CONTINUING PROJECTS**

Protocols for the sampling and experimentation on vertebrates in the 1995 proposal were reviewed and approved by the University of Alaska Institutional Animal Care and Use Committee. This assurance is valid for this proposal and has been reviewed and approved for renewal in FY 97. Current tests of oral dosing of harbor seals with <sup>15</sup>N-labeled amino acids have resulted in altered protocols at the Mystic Marinelife Aquarium. This revision will be incorporated into future protocols if the method is successful.

## **PROPOSED PRINCIPAL INVESTIGATOR**

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Voice: (907) 474-7115  
Fax: (907) 474-7204  
Email: [schell@ims.alaska.edu](mailto:schell@ims.alaska.edu)

## PRINCIPAL INVESTIGATOR

D. M. Schell has been involved in stable isotope studies for over 25 years. His research has included natural abundance tracer studies and enrichment experiments. His work on bowhead whales and geographic gradients in stable isotope ratios has been published and subjected to rigorous reviews. The findings have been upheld and have provided insight into bowhead whale natural history unattainable by other techniques.

D. M. Schell oversees the Stable Isotope Ratio Mass Spectrometry Facility at the University of Alaska Fairbanks campus. This consists of three working instruments which are dedicated to specific elements as demand requires. A Europa automated continuous flow system will be used for most samples but back-up analytical capability is available. A new automated machine has recently been funded by the National Science Foundation and ordered for delivery in July 1997. This machine will increase our sample analysis capability and provide greater sensitivity for small samples.

As PI, D. M. Schell will oversee the Quality Assurance/Quality Control aspects of this project. Protocols for sampling have been established and working standards are cross-calibrated with other nationally recognized laboratories.

## OTHER KEY PERSONNEL

Machine operations are the responsibility of Norma Haubenstock, technician. She is well trained and has more than nine years experience with mass spectrometers. Additional funds are budgeted for an assistant to prepare samples, load and operate the automated system and to aid in data processing and archiving for all users.

## LITERATURE CITED

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Michener, R.H. and D.M. Schell. 1994. The use of stable isotopes in tracing marine aquatic food webs. In: R. Michener and K. Lajtha (eds.), *Stable Isotopes in Ecology and Environmental Research*, pp. 138-157. Blackwell Scientific, Cambridge.

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Vinette, K.A. 1992. Carbon and nitrogen isotope ratios in bowhead whales and their zooplankton prey as indicators of feeding strategy and environmental change. M.S. Thesis, University of Alaska Fairbanks, Fairbanks, AK 147 pp.



# 1998 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1997 - September 30, 1998

Budget Category:	Authorized FY 1997	Proposed FY 1998						
Personnel	\$83.3	\$67.0						
Travel	\$11.0	\$6.3						
Contractual	\$5.7	\$4.0						
Commodities	\$7.1	\$5.1						
Equipment	\$0.0	\$0.0						
Subtotal	\$107.1	\$82.4	LONG RANGE FUNDING REQUIREMENTS					
Indirect	\$26.8	\$20.6	Estimated FY 1999	Estimated FY 2000	Estimated FY 2001	Estimated FY 2002	Estimated FY 2003	Estimated FY 2004
Project Total	\$133.9	\$103.0						
Full-time Equivalents (FTE)	1.3	1.0						
Other Funds			Dollar amounts are shown in thousands of dollars.					
Comments:  Indirect costs are 25% Total Direct Cost, the rate negotiated by the EVOS Trustee Council with the University of Alaska.								

**FY 98**

Project Number: 98170

Project Title: Isotope Ratio Studies of Marine Mammals in Prince  
William Sound

Name: University of Alaska Fairbanks

FORM 4A  
Non-Trustee  
SUMMARY

## COUNCIL PROJECT BUDGET

October 1, 1997 - September 30, 1998

Personnel Costs:			Months Budgeted	Monthly Costs	Overtime	Proposed FY 1998
Name	Position Description					
D. Schell	Professor – P.I.		2.3	11.7		26.3
N. Haubenstock	Technician – Mass Spectrometry		4.2	4.9		20.5
Vacant	Technician		6.0	3.4		20.2
Subtotal			12.5	20.0	0.0	
Personnel Total						\$67.0

Travel Costs:		Ticket Price	Round Trips	Total Days	Daily Per Diem	Proposed FY 1998
Description						
Fairbanks to Mystic, Connecticut -- live subject studies		1000	1	4	120	1.5
Fairbanks to Seward -- sampling		150	2	4	125	0.8
Fairbanks to Monaco (partial cost) -- present findings to SMM		1500	2	8	125	4.0
Travel Total						\$6.3

**FY 98**

Project Number: 98170  
Project Title: Isotope Ratio Studies of Marine Mammals in Prince  
William Sound  
Name: University of Alaska Fairbanks

FORM 4B  
Personnel  
& Travel  
DETAIL

**1998 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET**

October 1, 1997 - September 30, 1998

<b>Contractual Costs:</b>		Proposed
Description		FY 1998
Communications		0.7
Maintenance, mass spectrometry service		3.0
Shipping, expediting samples, equipment		0.3
<b>Contractual Total</b>		<b>\$4.0</b>
<b>Commodities Costs:</b>		Proposed
Description		FY 1998
Mass spectrometry-helium, high purity oxygen, liquid nitrogen		3.5
Chemicals, glassware, expendables		1.2
Project supplies (computer, printer supplies)		0.4
<b>Commodities Total</b>		<b>\$5.1</b>

**FY 98**

Project Number: 98170  
 Project Title: Isotope Ratio Studies of Marine Mammals in Prince  
                     William Sound  
 Name: University of Alaska Fairbanks

FORM 4B  
 Contractual &  
 Commodities  
 DETAIL

Prepared: 04/14/97

3 of 4

October 1, 1997.

ember 30, 1998

<b>New Equipment Purchases:</b>		<b>Number of Units</b>	<b>Unit Price</b>	<b>Proposed FY 1998</b>
<b>Description</b>				
Those purchases associated with replacement equipment should be indicated by placement of an R.		<b>New Equipment Total</b>		\$0.0
<b>Existing Equipment Usage:</b>			<b>Number of Units</b>	
<b>Description</b>				

**FY 98**

Project Number: 98170  
Project Title: Isotope Ratio Studies of Marine Mammals in Prince  
William Sound  
Name: University of Alaska Fairbanks

FORM 4B  
Equipment  
DETAIL



**KENAI HABITAT RESTORATION & RECREATION  
ENHANCEMENT PROJECT**

Project Number:	98180
Restoration Category:	Habitat Improvement
Proposer:	ADNR/ADFG
Lead Trustee Agency:	ADNR/ADFG
Cooperating Agencies:	DOI/USFS
Duration:	One Year
Cost FY 98:	\$864.4
Geographic Area:	Kenai Peninsula
Injured Resource/Service:	Pink salmon, sockeye salmon, Dolly Varden, commercial fishing, subsistence, recreation & tourism.

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EXXON VALDEZ OIL SPILL  
TRUSTEE COUNCIL

**ABSTRACT**

Adverse impacts to the banks of the Kenai River total approximately 19 miles of the river's 166 mile shoreline. Included in this total are 5.4 river miles of degraded shoreline on public land. Riparian habitats have been impacted by trampling, vegetation loss and structural development. This riparian zone provides important habitat for pink salmon, sockeye salmon and Dolly Varden, species injured by the *Exxon Valdez* oil spill. The project's objectives are to restore injured fish habitat, protect fish and wildlife habitat, enhance and direct recreation and preserve the values and biophysical functions that the riparian habitat contributes to the watershed. Restoration/enhancement techniques will include revegetation, streambank restoration, elevated boardwalks, floating docks, access stairs, fencing, signs, and educational interpretive displays.

## INTRODUCTION

This project is a continuation of the Kenai River Habitat Restoration and Recreation Enhancement Project that began in 1996. The objectives of this project are to:

1. Restore and protect fish habitat on the Kenai River,
2. Improve existing recreational access to the Kenai River watershed in a manner that restores and protects riparian fish and wildlife habitat,
3. Provide information to the public that promotes their understanding of the river's ecology and proper use of its resources.

Public lands on the Kenai Peninsula, including those soon to be acquired with *Exxon Valdez* oil spill joint settlement funds, contain important habitat for several species injured by the spill and provide recreation services for tens of thousands of Alaska residents and tourists. Kenai River fish support a large commercial fishery, a commercial sport fishing industry, a subsistence fishery, and a recreational sport fishery. In the aggregate, revenues generated by sportfishing, commercial fishing and river-based tourism represent a significant and growing proportion of the local economy.

The riparian zone, the transitional area that lies between the river's channel and the uplands, provides important fish and wildlife habitat and plays a major role in the hydrology of the watershed by helping to control floods and erosion. This vegetated area functions as a buffer and filter system between upland development and the river, thereby maintaining water quality by absorbing nutrients, accumulating and stabilizing sediments, and removing heavy metals and pollutants that are a result of urban development and which enter the river from surface runoff. It is also the area where a significant portion of the Kenai River's sportfishing and other recreational activities are concentrated.

Degradation of the river's streambanks, riparian vegetation and fish habitat has the potential of jeopardizing its long term productivity and degrading the quality of the recreational experience. This project proposes revegetation, streambank restoration, and public access improvements that will promote pink and sockeye salmon and Dolly Varden habitat protection and restoration, as well as enhancement of recreational services in the Kenai River watershed. The project also proposes to design and construct educational and interpretive displays that will inform the public of the proper manner in which to access and use the river's resources.

During 1996, the following project elements were accomplished:

1. Development of site assessment and nomination procedures,
2. Development of a digital database containing site assessment and nomination data,
3. Development of an evaluation and ranking process for nominated projects,

4. An Interdisciplinary Team (IDT) of biologists, resource managers and planners was selected to review evaluation procedures and nominated projects,
5. Review and evaluation, by the IDT, of 16 projects nominated by public landowners,
6. Public scoping meetings were held in Anchorage, Kenai and Cooper Landing to discuss the project,
7. Production and publication of an Environmental Assessment (EA) document,
8. Review and response to EA comments,
9. Development of Cooperative Agreements that will form the basis for funding projects carried out by public landowners,
10. Consummation of agreements between ADF&G/ADNR and public landowners for five projects will take place in 1997.

During 1997, the following project elements were accomplished:

1. Review and evaluation, by the IDT, of 7 new projects nominated by public landowners,
2. Production and publication of an Environmental Assessment (EA) document for the 1997 projects,
3. Oversight and field inspections of Kenai Beach Dunes project.

Restoration and enhancement proposals on public lands extending from the outlet of Kenai Lake to the mouth of the Kenai River (Figure 1), were nominated by public landowners and evaluated by an Interdisciplinary Team (IDT) of biologists and resource managers using specific threshold and evaluation criteria (Table 1). The IDT designed the qualifying criteria used to evaluate and rank the proposals by considering a variety of factors, including the degree of damage at a site and the effects that each proposal will have on fish habitat, recreation, and the surrounding environment.

Conceptual restoration and enhancement plans were presented to the IDT for evaluation. Final engineered plans will be provided to ADFG/ADNR prior to construction. Choice of building materials and construction methods are the responsibility of the landowner (but subject to IDT review) and must employ restoration techniques permissible by regulatory agencies (ADFG, ADNR, and the Army Corps of Engineers).

The project was proposed to last for three years, beginning in 1996. The seven qualifying proposals initiated in 1996 will be completed in 1997. Construction was started on one in 1996 and construction will begin on the other six this spring. Projects approved for funding in 1997 will be completed in 1998. Monitoring of funded proposals will be carried out by ADFG/ADNR to ensure the proposals are constructed and function as designed.



Monitoring will also be used to gather information regarding effectiveness of restoration techniques.

Seven proposals (Table 2) were evaluated and scored according to threshold and evaluation criteria. One proposal, Kenai Mouth-South Side Access, was disqualified because it did not fulfill all threshold criteria. The majority of funding for the Centennial Park project will come from another source. If funding is approved, six sites will be restored in 1998.

Because all proposals had to meet threshold criteria before the evaluation criteria were applied, six proposals are eligible for funding. The scores are a method of ranking those proposals that best achieve the overall project's goals for habitat restoration, compatible recreation enhancement, and educational value. In an attempt to identify the most cost-effective proposals and obtain maximum benefits from available funds, it was decided to compare the relative restoration benefits of the proposals in terms of costs. To facilitate that determination, the results of the evaluation process, i.e. the scores, were plotted against the estimated costs. Figure 2 displays the relative or comparative restoration benefits of the 1997 proposals as a function of cost. Figure 3 is a composite plot of the 1996 and 1997 nominations.

Cooperative agreements or Reciprocal Service Agreements (RSA's) will be negotiated and signed for the projects identified in the Preferred Alternative of the EA. Construction should begin on these five proposals in 1997 and 1998.

Work proposed for 1998 includes:

1. Oversight and monitoring of on-going projects,
2. Finalizing cooperative agreements with public landowners for projects to be constructed in 1998,
3. Review and evaluation of new nominations for projects on other public lands,
4. Design and development of educational and interpretive materials,
5. Preparation of an annual report.

**Table 1: Threshold and Evaluation Criteria**

**Threshold Criteria**

1. The project will protect, restore or enhance the historic functional attributes of a site and the surrounding area.
2. The project is located on public land.
3. The managing agency agrees to endorse the project.
4. The managing agency agrees to future maintenance and management of the project in a manner that facilitates and is consistent with the restoration or enhancement endpoint (#1).
5. All elements of the project can be permitted.
6. The project is not a mitigation requirement.

**Nomination must be in compliance with all Threshold Criteria.**

**Evaluation Criteria**

1. Potential Habitat Value  
What is the potential habitat value of the project? [Score =  $(20/10/5) \times 3.5$ ]
2. Potential Recreation Value  
What is the potential recreation value of the project? [Score =  $(20/10/5) \times 2.5$ ]
3. Disturbance Level  
What is the level of disturbance (human impact) in relation to habitat/recreation values?  
[Score =  $(20/10/5) \times 2.0$ ]
4. Rate  
To what extent will the project decrease the amount of time needed for riparian habitat to recover? [Score =  $(20/10/5) \times 1.0$ ]
5. Collateral Impacts  
What is the potential for adverse impacts to natural or cultural resources or to the nearby human community resulting from this project?  
[Inverse relationship: Score =  $(5/10/20) \times 3.0$ ]
6. Design/Effectiveness  
How would you rate the project's design to its expected effectiveness?  
[Score =  $(20/10/5) \times 2.0$ ]
7. Vulnerability  
Is the protected, restored or enhanced site vulnerable to natural or human-induced degradation. [Inverse relationship: Score =  $(5/10/20) \times 2.0$ ]

**Table 2: 1997 Project Evaluation Summary**

<u>Project ID</u>	<u>Project Name</u>	<u>Project Score</u>
K 17	Cone	222
K 18	Kobylarz	253
K 19	Russian River Phase 2	241
K 20	Centennial Park 97	294
K 21	Slikok Creek	300
K 22	Bing's Landing	261

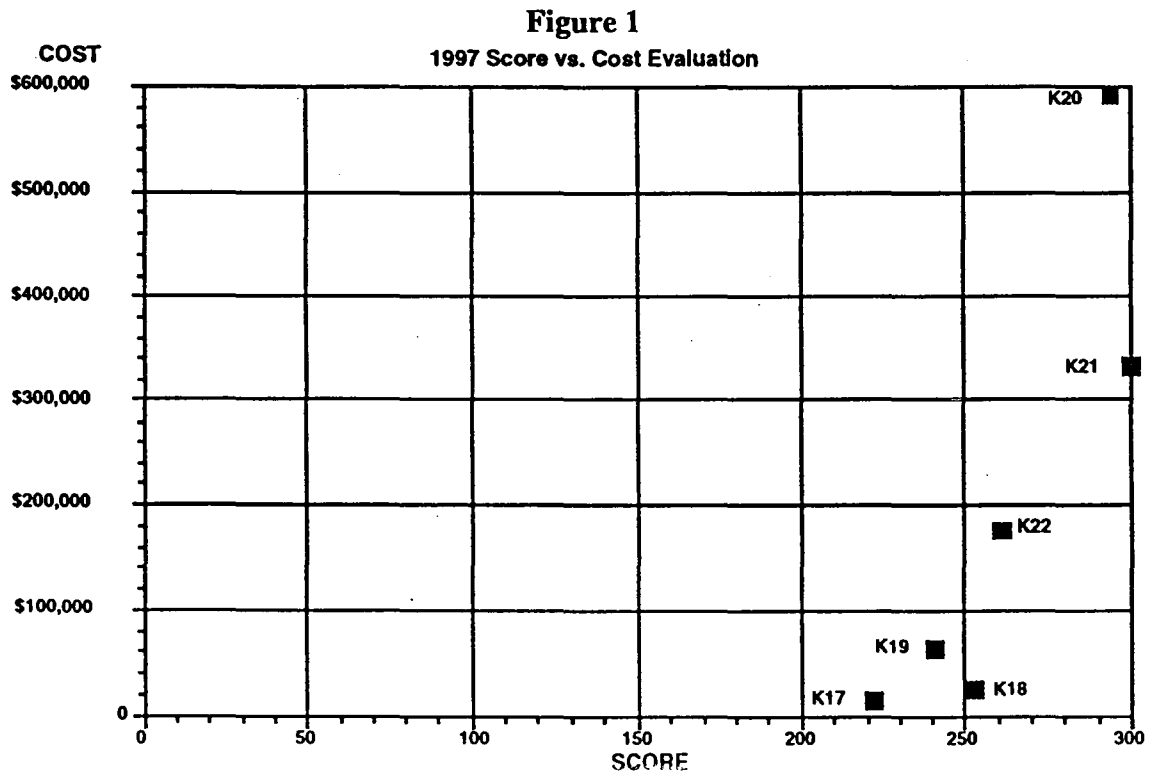
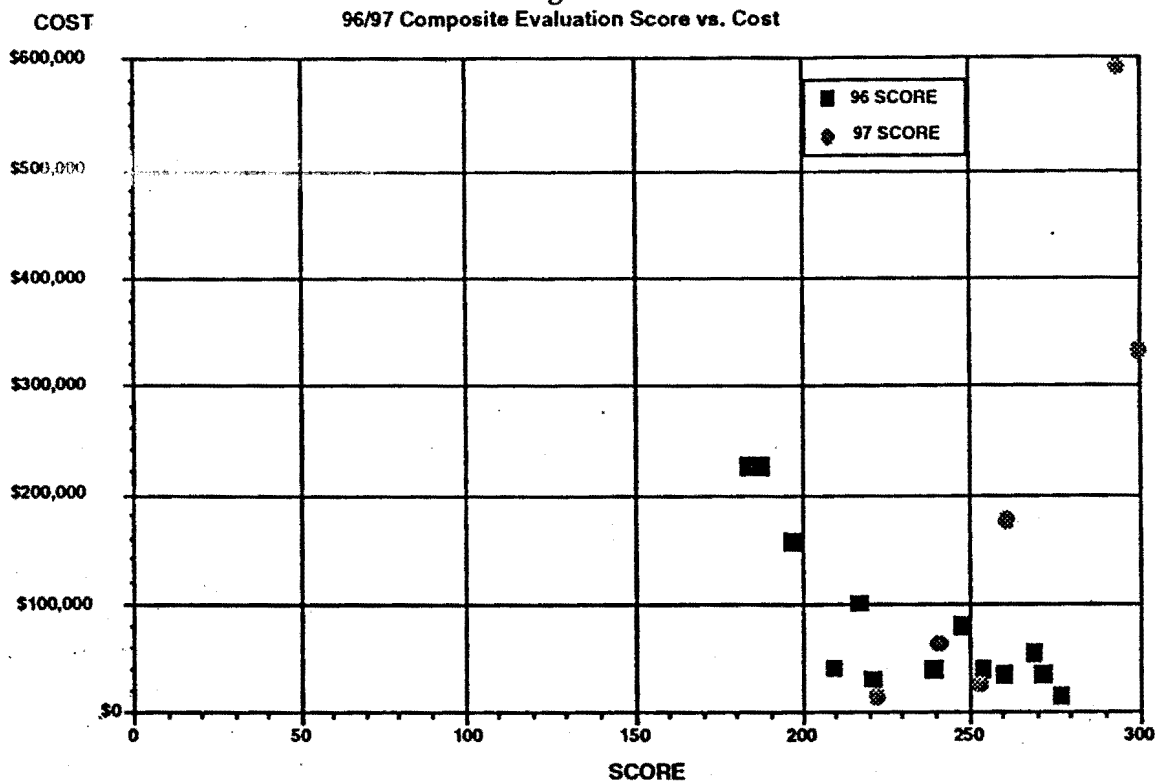


Figure 2:



## NEED FOR THE PROJECT

### A. Statement of Problem

Use of the Kenai River watershed is degrading fish habitat along the riparian zone of the mainstem and, to a lesser degree, the tributaries of the river. Streambanks that provide essential fish habitat are being trampled and denuded of vegetation leading to increasing rates of erosion and sedimentation. Both commercial and residential developments are altering shorelines, changing patterns of runoff and creating the potential for the discharge of non-point source pollutants into the river. Federal and state resource agencies have limited ability to manage these problems that have the potential of threatening the productivity and world class recreational value of this river system.

Commercial fishing, subsistence, recreation and tourism (including sport fishing) are services that were reduced or lost because of the spill. Within the Kenai River watershed, the resources that support these services that were injured by the *Exxon Valdez* oil spill include pink and sockeye salmon and Dolly Varden. Chinook and coho salmon also contribute significantly to these services. The *Exxon Valdez* Oil Spill Restoration Plan states that the Kenai River sockeye salmon population is not recovering and that: *With regard to sockeye salmon, the objective of habitat protection is to ensure maintenance of adequate water quality, riparian habitat, and intertidal habitat.*

The restoration strategy articulated in the restoration plan for recreation and tourism focuses on the: *Preservation and improvement of the recreational and tourism values of the spill area*. The Plan goes on to discuss strategies for promoting recovery of commercial fishing, recreation and tourism by: *...increasing the availability, reliability, or quality of the resource on which the service depends*.

What is needed within the Kenai River watershed is an integrated approach that protects resource habitats, restores degraded streambanks and riparian vegetation, maintains productivity and promotes appropriate, sustained human use of the river.

## **B. Rationale**

The work proposed by this project is needed to protect and restore fishery resources. Continuing loss of habitat will exacerbate the injury caused by the spill to both resources and services and lead to diminished productivity. This, in turn, diminishes the value of the commercial, subsistence and sport fisheries and the quality of recreation on the river with significant, adverse implications for the local economy.

Based on a review of historic recreation use patterns and habitat impacts, the project will protect, restore, stabilize, or rehabilitate streambanks where resource damage is occurring; enhance or close existing access points and movement corridors; or re-direct users to other areas of the river on a temporary or long term basis. These actions will be based on the need to facilitate human use of the river in a way that protects fish habitat and minimizes degradation of other sensitive and/or pristine habitats.

This project is designed to promote streambank stability, increase vegetative cover, and mitigate accelerated erosion and sedimentation for the benefit of pink salmon, sockeye salmon, Dolly Varden and other fish species that migrate and rear along the river's banks. Techniques used to achieve these goals may include the use of elevated, grated boardwalks, river access stairs, fishing platforms, spruce tree revetments and other riparian habitat improvement and protection techniques. These techniques will, at the same time, restore and enhance sportfishing. One example is elevated, grated boardwalks, constructed to protect revegetating streambanks, that will provide river access to anglers with a minimum of impact to the recovering habitat. Post-construction monitoring will examine the effects of the method and the amount of recreational use that occurs in the area.

The education component of the project will produce user information and interpretive displays at strategically located access points along the river. These displays will provide users with information on the natural history of the river's fish, their habitats, ecology of the river system and the best methods that they can use to maximize their recreational experience with a minimum of impact to the watershed and its resources. Signs placed adjacent to work sites will describe the on-going restoration effort and direct the public away from recovering vegetation.

Each site under consideration for a restoration, enhancement or education project will be evaluated in terms of the condition of its habitats, character of adjacent lands, and historic public use. Improvements to access will reflect patterns of use as well as on-site and adjacent upland environmental sensitivities.

### **C. Location**

All construction, maintenance and monitoring components of the project will be located within the Kenai River watershed. Planning and coordination will be based in Anchorage. Primary ecological benefits from the project will be realized by the natural systems within the watershed. Secondary benefits will affect the economy of the communities of the Kenai Peninsula and the commercial fishing industry. Improved and enhanced recreation benefits will affect users from southcentral Alaska as well as tourists from outside of the state. Communities that may be affected by the project include: Kenai, Soldotna, Homer, Sterling, Cooper Landing, Anchorage and the unincorporated communities on the Kenai Peninsula.

### **COMMUNITY INVOLVEMENT**

It is intended that the project be fully integrated with on-going agency recreation management, permitting and regional planning activities affecting the Kenai River watershed. This includes coordination with the Kenai Peninsula Borough, City of Kenai, Kenai City Council, City of Soldotna, Soldotna City Council, Kenai Peninsula Borough Assembly, and local interest groups.

### **PROJECT DESIGN**

#### **A. Objectives**

1. Solicit restoration project nominations from public land managers on the Kenai River.
2. Evaluate and rank projects on the basis of their restoration benefit and cost effectiveness.
3. Review detailed design plans and develop cooperative agreements for construction of the projects.
4. Verify compliance with restoration designs and evaluate construction.
5. Implement a monitoring program to assess restoration and use of project sites.
6. Design and construct educational and interpretive signs and displays.

#### **B. Methods**

The present condition of North America's native fish fauna is attributable, in part, to the degradation of aquatic ecosystems and habitat (FEMAT Report, 1993). Loss and degradation of freshwater habitats are the most frequent factors responsible for the decline of anadromous salmonid stocks (Nehlsen, et. al. 1991). Along with habitat modification

or loss, changes in water quality and quantity are often cited as causative factors for degradation of aquatic systems and declines in anadromous fish populations.

The Kenai River Cumulative Impacts Assessment of Development Impacts on Fish Habitat (Liepitz, 1994) was designed to identify and evaluate the cumulative impacts of development actions including public and private land use impacts on Kenai River fish habitat. The study documented that : 11.1 percent to 12.4 percent (18.4 to 20.6 miles) of the river's 134 miles of upland and 32 miles of island shoreline and nearshore habitats have been impacted by bank trampling, vegetation denuding, and structural development along the river's banks. Degraded public land along the Kenai River includes 5.4 miles of trampled riparian habitat and 3.5 miles of developed shoreline.

Site specific project designs will reflect site characteristics including: topography, hydrologic variables, vegetation, soils, extent and type of degradation and historic use patterns. Designs may include elements that restore or enhance specific habitat values. For example, instream structures may be used to enhance fish habitat and/or angler access. Plant propagation and streambank restoration techniques will be selected on the basis of site characteristics, constraints and cost. Revegetation designs will attempt to re-establish the native, riparian plant communities. Grasses that have been successfully used for riparian and saltmarsh revegetation in Alaska include: bluejoint reedgrass (*Calamagrostis canadensis*), Bering hairgrass (*Deschamsia beringensa*), sloughgrass (*Beckmannia syzigachne*), sedges (*Carex* spp.) and beach wildrye (*Elymus mollis*).

Successful revegetation requires control of site impacts. Consequently, fences and/or signed closures may be required to protect undamaged sites from human impact or to prevent additional damage to recovering sites. Project areas will either be closed and posted during the course of revegetation, or environmental engineering techniques will be used that allow public access but protect the recovering habitat from additional adverse impacts. Habitat improvement and protection techniques to be considered include:

On-site Revegetation/Restoration	Signage
Exclosures	Elevated Grating/Boardwalks
Spruce Tree Revetments	Access Stairs Ladder
Access Trails	Floating Docks

The number of sites selected for revegetation or enhancement in a given year will be dependent upon the time necessary for completion, i.e., permitting, construction and installation, and the availability of funding.

Educational/interpretive displays will be designed, constructed and placed in strategic locations along the river. Signs will also be designed and located to prevent bank trampling in areas where revegetation efforts are occurring.

A monitoring program will be used to evaluate the degree of success of each project. The purpose of the monitoring program is to:

1. Determine if the project is in compliance with the Cooperative Agreement.
2. Evaluate whether the project was been successful in meeting the restoration goals set forth in the project description, and
3. Provide data that will help in design of future restoration projects and in the establishment of performance standards.

Monitoring parameters will be chosen that reflect site-specific restoration/enhancement objectives and may include habitat, vegetation and public use measurements. The assessment of the existing condition of each site will serve as the baseline for monitoring. Monitoring measurements will be obtained frequently early in the project and could be used to amend the design if necessary. Wherever possible, photo plots will be installed and photos taken biannually. Once the project is successfully constructed and it is determined that restoration/enhancement is proceeding on an acceptable course and rate, monitoring measurements will be taken less frequently. Projects that are initially monitored monthly during the early stages of vegetation growth and establishment will be monitored biannually thereafter. Habitat and population monitoring parameters may include: vegetation diversity and cover, fish utilization and stream stability. Public use of the sites and impacts to adjacent areas will also be monitored. Site visitation shall be based on counts of individual people by field staff and project personnel.

Observations may be made during winter months to evaluate the effects of ice scouring. The period that a project is monitored will be based upon the amount of time required for achievement of objectives.

#### **C. Cooperating Agencies, Contracts and Other Agency Assistance**

All components of the project will be carried out by personnel from ADF&G and ADNR. Volunteers supervised by agency staff will assist in the installation of prefabricated structures and in routine maintenance. Cooperating agencies will participate in IDT evaluations and development of a supplement EA. Coordination will occur with agencies through contract administration and oversight.

### **SCHEDULE**

#### **A. Measurable Project Tasks for FY 98**

October 1 to December 1:     Contract administration.  
                                      Project monitoring.  
                                      Preparation of annual report.



December 1 to May 15:	Review detailed design plans. Design and produce educational materials and signs. Establish cooperative agreements with public landowners for second round.
May 15 to July 15:	Management and oversight of project construction. Contract administration. Put up signs and information displays.
July 15 to August 15:	Inspect all project sites to check for compliance with design parameters. Monitor revegetation sites. Monitor public use of completed project and proposed sites for next year.
August 15 to Sept. 30:	Continue monitoring. Contract administration.

#### **B. Project Milestones and Endpoints**

Oct. 1--Nov.1:	Complete construction on all projects. Inspect the projects to check for compliance with design and construction parameters Close-out completed cooperative agreements
Feb1--May 15:	Publish supplemental EA Consummate cooperative agreements with public landowners for third round projects
July 15 to Sept. 30:	Complete summer monitoring and project compliance inspections

#### **NORMAL AGENCY MANAGEMENT**

The impacts affecting the Kenai River are occurring at a rate and magnitude far in excess of the management resources that are available to mitigate or restore habitat damage. The proposed project supplements existing efforts to reverse this trend. Moreover, none of the riparian habitat on small parcels that the Trustee Council is acquiring on the Kenai River has been surveyed or evaluated for restoration work. Additional issues relevant to state agency management of the Kenai River are to be found in the following section.

## COORDINATION AND INTEGRATION OF RESTORATION EFFORT

Coordination will occur with agency staffs in DNR, ADF&G and the Kenai National Wildlife Refuge. Their expertise will be used in defining management objectives, developing criteria, evaluating and ranking potential project sites, conducting archaeological and historical reviews and clearances, performing design to include preparing plans and specifications, bidding construction projects, oversight of project construction, permitting, monitoring public use, and enforcing site restrictions.

The project will build upon pilot efforts that have been implemented or are being developed for the river. In 1994, boardwalks were installed near the Soldotna airport and on numerous private parcels; exclosures have been used with a high degree of success along portions of the Russian River and in units of the state park system. State permitting procedures have also resulted in numerous bank stabilization projects that maintain or enhance fish habitat by using spruce tree revetments, root wads, live willow cuttings, and other protective measures.

The state and federal governments have already committed funds to accomplish several of the objectives identified by this project. Fish and Game *Exxon Valdez* criminal settlement funds (\$3 million) have been dedicated for the construction of habitat protection demonstration projects and land acquisition on the Kenai River. The U.S. Fish and Wildlife Service has provided challenge grant funding to assist the ADF&G demonstration projects. The National Marine Fisheries Service will provide the ADF&G with an additional one million dollars for streambank improvements under an appropriation requested by Senator Stevens. ADNR restitution funds (\$7 million) will be used, in part, to construct boardwalks and access platforms that protect streambanks at heavily used state park units at Morgan's Landing, Bing's Landing, and Slikok Creek. Dingle-Johnson funds are being used to provide recreational access, streambank revegetation, and streambank protection structures at The Pillars project site.

The intense public use pressures and development activities on the Kenai River threaten to overwhelm the limited budgets available to resource agencies attempting to manage the river for resource protection and sustained recreational use. That is why supplementary funding is so important. The proposed project, along with those utilizing other available funds, provides a cost-effective method to protect streambanks and minimize further habitat degradation.

## EXPLANATION OF CHANGES IN CONTINUING PROJECTS

The project design and schedule described in the DPD approved by the Trustee Council for FY96 and FY 97 are unchanged.

## REFERENCES

Alaska Dept. of Fish and Game and Alaska Dept. of Natural Resources. 1986. Field Guide for Streambank Revegetation . Anchorage, AK: Alaska Dept. of Fish and Game.

Forest Ecosystem Management Team. 1993. Forest Ecosystem Management: An Ecological, Economic, and Social Assessment. Portland, OR: U.S. Forest Service. FEMAT Report.

Kondolf, G.M. 1995. Five elements for effective evaluation of stream restoration. *Restoration Ecology* 3:133-136.

Liepitz, Gary S. 1994. An Assessment of the Cumulative Impacts of Development and Human Uses on Fish Habitat in the Kenai River. Anchorage, AK: Alaska Department of Fish and Game, Habitat and Restoration Division. Technical Report No. 94-6.

Schiechtl, Hugo. 1980. Bioengineering for Land Reclamation and Conservation. 404 pgs. Edmonton, Alberta: Univ. Alberta Press.

Sherman, Jensen E. and William, S. Platts. 1990. Restoration of Degraded Riverine/Riparian Habitat in the Great Basin and Snake River Regions. *Wetland Creation and Restoration*, p. 367-404. Kusler, Jon, A. and Mary E. Kentula ed. Washington, D.C.: Island Press.

Sowl, John H. 1990. Restoration of Riparian Wetlands Along a Channelized River: Oxbow Lakes and the Middle Missouri. *Environmental Restoration*, p. 294-305. Berger, John, J. ed. Washington, D.C.: Island Press.

## PERSONNEL

### Project Leader

Mark Kuwada - Habitat Biologist with the Alaska Department of Fish and Game for 15 years. Extensive experience in coordinating departmental policy and mitigating major project impacts; Project Manager for Federal OCS Oil and Gas Leasing Program; Susitna Hydroelectric Project; Bradley Lake Hydroelectric Project; Diamond Chuitna Coal Project; ADF&G Response Coordinator, *Exxon Valdez* oil spill. ADF&G Title 16 permitter for southcentral Alaska and the Kenai River.

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### Project Leader

Art Weiner, Ph.D - Natural Resources Manager with the Alaska Department of Natural Resources for seven years. B.S., M.S. and Ph.D in biology. Extensive experience in field biology, permitting, design and construction of restoration projects and in coordinating department policy with other state and federal resource agencies.

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FAX (907) 269-8902

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### Project Leader

# 1997 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1996 - September 30, 1997

Budget Category:	Authorized FFY 1997	Proposed FFY 1998	PROPOSED FFY 1997 TRUSTEE AGENCIES TOTALS					
			ADEC	ADF&G	ADNR	USFS	DOI	NOAA
				\$308.7	\$473.5			
Personnel	\$168.0	\$167.5						
Travel	\$9.5	\$10.4						
Contractual	\$357.6	\$621.3						
Commodities	\$14.0	\$2.7						
Equipment	\$0.0	\$0.0	LONG RANGE FUNDING REQUIREMENTS					
Subtotal	\$549.1	\$801.9	Estimated FFY 1999	Estimated FFY 2000	Estimated FFY 2001	Estimated	Estimated	
General Administration	\$50.3	\$62.5						
Project Total	\$599.4	\$864.4	\$0.0	\$0.0	\$0.0	\$0.0	\$0.0	\$0.0
Full-time Equivalents (FTE)	0	2.1						
			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: Projects will be funded at an amount not to exceed the amount proposed as the project was initially evaluated. Individual project budget detail is being developed as a part of cooperative agreement with the managing entity.								

1998

Project Number: 98180  
Project Title: Kenai Habitat Restoration and Recreation Enhancement  
Lead Agency: AK Dept. of Natural Resources

FORM 2A  
MULTI-TRUSTEE  
AGENCY  
SUMMARY

Prepared:

**1997 EXXON VALDEZ TRUST\_\_\_ \_OUNCIL PROJECT BUDGET**

October 1, 1996 - September 30, 1997

Budget Category:	Authorized FFY 1996	Proposed FFY 1997						
Personnel	\$84.0	\$84.0						
Travel	\$4.8	\$4.8						
Contractual	\$206.5	\$352.4						
Commodities	\$8.5	\$0.2						
Equipment	\$0.0	\$0.0	LONG RANGE FUNDING REQUIREMENTS					
Subtotal	\$303.8	\$441.4	Estimated FFY 1999	Estimated FFY 2000	Estimated FFY 2001	Estimated	Estimated	
General Administration	\$27.1	\$32.1						
Project Total	\$330.9	\$473.5						
Full-time Equivalents (FTE)		1.0						
Dollar amounts are shown in thousands of dollars.								
Other Resources								
Comments:								

**1998**

Prepared:

Project Number: 98180  
 Project Title: Kenai Habitat Restoration and Recreation Enhancement  
 Agency: AK Dept. of Natural Resources

FORM 3A  
 TRUSTEE  
 AGENCY  
 SUMMARY

**October 1, 1996 - September 30, 1997**

<p><b>1998</b></p>	<p>Project Number: 98180          Project Title: Kenai Habitat Restoration and Recreation Enhancement          Agency: AK Dept. of Natural Resources</p>	<p>FORM 3B          Personnel          &amp; Travel          DETAIL</p>
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4/14/97

**1997 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET**  
 October 1, 1996 - September 30, 1997

<b>Contractual Costs:</b>		<b>Proposed</b>
<b>Description</b>		<b>FFY 1998</b>
Cooperative Agreements for the following projects:		
Slikok Creek		\$332.4
Signage		\$20.0
Map production for presentation and signage.		
When a non-trustee organization is used, the form 4A is required.		
<b>Contractual Total</b>		<b>\$352.4</b>
<b>Commodities Costs:</b>		<b>Proposed</b>
<b>Description</b>		<b>FFY 1998</b>
Office supplies (including paper, toner cartridges, data cartridges, mailing labels, write in rain paper, etc.)		0.2
<b>Commodities Total</b>		<b>\$0.2</b>

**1998**

Prepared:

Project Number: 98180  
 Project Title: Kenai Habitat Restoration and Recreation Enhancement  
 Agency: AK Dept. of Natural Resources

FORM 3B  
 Contractual &  
 Commodities  
 DETAIL



**October 1, 1996 - September 30, 1997**

1998

Project Number: 98180  
Project Title: Kenai Habitat Restoration and Recreation Enhancement  
Agency: AK Dept. of Natural Resources

**FORM 3B**  
**Equipment**  
**DETAIL**

5 of 17

4/14/97

**1997 EXXON VALDEZ TRUST \_ \_ JUNCIL PROJECT BUDGET**

October 1, 1996 - September 30, 1997

<b>Budget Category:</b>	<b>Authorized FFY 1997</b>	<b>Proposed FFY 1998</b>							
Personnel	\$84.0	\$83.5							
Travel	\$4.7	\$5.6							
Contractual	\$151.1	\$268.9							
Commodities	\$5.5	\$2.5							
Equipment	\$0.0	\$0.0							
Subtotal	\$245.3	\$360.5	LONG RANGE FUNDING REQUIREMENTS						
General Administration	\$23.2	\$30.4	Estimated FFY 1999	Estimated FFY 2000	Estimated FFY 2001	Estimated	Estimated		
Project Total	\$268.5	\$390.9							
Full-time Equivalents (FTE)		1.1							
Other Resources			Dollar amounts are shown in thousands of dollars.						
Comments:									

**1998**

Project Number: 98180  
 Project Title: Kenai Habitat Restoration and Recreation Enhancement  
 Agency: AK Dept. of Fish & Game

FORM 3A  
 TRUSTEE  
 AGENCY  
 SUMMARY

Prepared:

October 1, 1996 - September 30, 1997

1998

Project Number: 98180  
Project Title: Kenai Habitat Restoration and Recreation Enhancement  
Agency: AK Dept. of Fish & Game

FORM 3B  
Personnel  
& Travel  
DETAIL

Prepared:

**1997 EXXON VALDEZ TRUST      COUNCIL PROJECT BUDGET**  
October 1, 1996 - September 30, 1997

<b>Contractual Costs:</b>		Proposed
Description		FFY 1998
Signage		20.0
Cooperative Agreements for:		
Cone		15.0
Kobylarz		25.0
Russian River, Phase II		63.9
Centennial Park Angler Trail		45.0
Russian River, Phase III		100.0
When a non-trustee organization is used, the form 4A is required.		
<b>Contractual Total</b>		<b>\$268.9</b>
<b>Commodities Costs:</b>		Proposed
Description		FFY 1998
Field equipment as needed for project oversight and monitoring.		2.0
Office supplies (including paper, toner cartridges, data cartridges, mailing labels, write in rain paper, etc.)		0.5
<b>Commodities Total</b>		<b>\$2.5</b>

**1998**

Prepared:

Project Number: 98180  
Project Title: Kenai Habitat Restoration and Recreation Enhancement  
Agency: AK Dept. of Fish & Game

**FORM 3B**  
**Contractual &**  
**Commodities**  
**DETAIL**

**1997 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET**  
October 1, 1996 - September 30, 1997

[illegible]

## 1998

Project Number: 98180  
Project Title: Kenai Habitat Restoration and Recreation Enhancement  
Agency: AK Dept. of Fish & Game

FORM 3B  
Equipment  
DETAIL

**Prepared:**



## Coded Wire Tag Recoveries From Pink Salmon in Prince William Sound

Project Number: 98186  
Restoration Category: General Restoration and Research/Monitoring  
Proposer: Alaska Department of Fish and Game  
Lead Trustee Agency: Alaska Department of Fish and Game  
Cooperating Parties: Prince William Sound Aquaculture Corporation  
Valdez Fisheries Development Association  
Alaska SeaLife Center:  
Duration: Tenth year, eleven year project  
Cost FY 98: \$ 126.6 Close-out  
Cost FY 99: \$0  
Cost FY 00: \$0  
Cost FY 01: \$0  
Cost FY 02: \$0  
Geographic Area: Prince William Sound  
Injured Resource/Service: Pink Salmon

RECEIVED  
APR 15 1997  
EXXON VALDEZ OIL SPILL  
TRUSTEE COUNCIL

### ABSTRACT

Pink salmon play a major role in the Prince William Sound ecosystem as well as in the economy of Cordova and other local communities. There is evidence that the *Exxon Valdez* oil spill has been partially responsible for weak pink salmon returns. Pink salmon runs are dominated by hatchery populations, and efforts to restore injured wild populations through selective harvesting of hatchery fish depend upon the availability of data pertaining to the spatial and temporal abundance of wild fish in different fishing areas. This study provided real-time and post-season estimates of hatchery and wild contributions to commercial harvests by date and fishing district, and also to hatchery cost-recovery harvests. This information enabled fisheries managers to direct effort away from injured populations.

## INTRODUCTION

Pink salmon play a major role in the Prince William Sound ecosystem. Migrating pink salmon fry are an important spring food source for various fish, birds and terrestrial mammals. Marine mammals, birds, and fish also prey on the ocean life stages of pink salmon and returning adult wild salmon comprise a large portion of the summer diet of terrestrial mammals and birds such as bears, river otters, wolverines, bald eagles, gulls, and kittiwakes. Returning adult salmon also provide a pathway for the transfer of nutrients accumulated from high seas marine areas to near-shore and terrestrial ecosystems. As the principal species harvested in the Prince William Sound salmon purse seine fishery, pink salmon play a major role in the commercial fishing and fish processing industries which are the backbone of the economy in Cordova and other Prince William Sound communities. Ex-vessel values for this fishery ranged from \$10 to almost \$40 million through the 1980's.

Prince William Sound pink salmon returns originating from brood years subsequent to the March 24, 1989, *T/V Exxon Valdez* oil spill have been aberrant or weak, with the exception of those of 1994. Returns of wild and hatchery pink salmon in 1991 arrived late, had very compressed run timing, and the fish were small and of poor commercial quality. Returns of pink salmon in 1992 and 1993 were far fewer than expected, while those of 1994, 1995 and 1996 were more in line with expectations. The 1992 return of wild pink salmon was the fourth smallest even year return in the last 30 years and the hatchery return was less than one third of expected. The 1993 return of wild pink salmon was the third smallest in the last 30 years and the hatchery return was less than one fifth of expected. Both wild and hatchery returns of 1994, 1995, and 1996 were a significant improvement over the preceding two years.

There is a growing body of evidence which indicates that the *Exxon Valdez* oil spill was partially responsible for the weak pink salmon returns to Prince William Sound. Much of the spawning for wild pink salmon (up to 75% in some years) occurs in intertidal areas. Intertidal spawning areas are susceptible to marine contaminants and there is strong evidence the *Exxon Valdez* oil spill adversely affected spawning success and early marine survival in Prince William Sound. Mortalities of pink salmon embryos incubating in the intertidal portions of oiled streams in western Prince William Sound have been significantly higher than those of embryos which incubated in nearby unoiled streams (Sharr et. al. 1994a, Bue et al. 1996). Despite apparent reductions in the amount of observable oil in intertidal salmon spawning areas since 1990, the differences in mortality between oiled and unoiled streams persisted in 1991, 1992 and 1993 and were also observed in spawning areas upstream of oil influence (Sharr et. al. 1994b, Bue et al. 1996). These findings may be indicative of heritable genetic damage which may have resulted in reproductive impairment among first and second generation fish originating from populations which incubated in oiled streams in 1989 and 1990.

In addition to damage incurred during the embryo stages of development, pink salmon fry and juveniles rearing in the western portions of Prince William Sound in 1989 also exhibited reduced



growth and survival (Willette and Carpenter, 1994). Because almost all wild and hatchery fry exit Prince William Sound through the straits and passages that were most heavily oiled, it is likely that at least portions of almost all pink salmon populations in Prince William Sound were damaged as rearing fry and juveniles in 1989. There are presently no data to substantiate any hypotheses regarding heritable damage to populations which traveled and fed in oiled marine waters as fry in 1989. Nevertheless, such a possibility is plausible given the findings of Sharr et al. (1994c).

Although hatchery pink salmon production in Prince William Sound began in the 1970's, the large returns associated with maximum permitted fry production did not occur until the late 1980's and early 1990's and consequently coincided with the *Exxon Valdez* oil spill era. Returns of wild salmon are dominated by the more productive hatchery populations and are therefore heavily exploited in commercial, sport, and subsistence fisheries. To sustain production from wild populations, managers must insure adequate escapements of wild fish to their natal streams, and that the escapement occurs in a smooth fashion over the season so that the genetic make-up of the populations is maintained. To achieve these goals, mixed-stock fisheries must be managed to achieve exploitation rates appropriate for the less productive wild populations throughout the season. Managers need, therefore, to be able to estimate the relative spatial and temporal abundance of wild fish in the different fishing areas of Prince William Sound.

The coded wire tag study provided real-time and post-season estimates of hatchery and wild contributions to commercial and hatchery cost-recovery harvests by date and fishing district. Such catch contribution estimates, together with real-time escapement estimates from an Alaska Department of Fish and Game aerial survey program were used inseason by fisheries managers to reduce exploitation of wild stocks and to target effort on hatchery returns. Post season analyses of tag recovery data coupled with escapement data for wild populations permitted estimation of total wild returns, which in turn allowed assessment of the effectiveness of various management strategies. Post season analyses also identified time and area distribution trends for wild and hatchery fish in fisheries. This information is important for fisheries managers who must anticipate the effects of fishing strategies in future years if injured populations are to be protected. Restoration processes have been used to justify time and area fishery closures and effectively reduce exploitation on oiled populations in portions of southwestern Prince William Sound in 1990 through 1996..

The results of the coded wire tag recovery project were also critical to the success of an integrated package of the Sound Ecosystem Assessment program. The Sound Ecosystem Assessment proposal has roots in a broader plan developed by the Prince William Sound Fisheries Ecosystem Research Planning Group, a bioregional coalition of Prince William Sound scientists, resource managers, resource users, aquaculture associations, and communities, formed to "develop an ecosystem level understanding of the natural and man-caused factors influencing the production of pink salmon...in Prince William Sound". Many of the Sound Ecosystem Assessment program projects, such as those falling under the Salmon Growth Component and

the Salmon Predation Component were dependent upon information provided by this coded wire tag study.

In the absence of the improved management capabilities afforded by this project, salmon stocks in western Prince William Sound which have been injured and depleted through oil impacts would have been put at greater risk in the commercial, sport and subsistence fisheries. Population levels of stocks might have been reduced below those needed for rapid recovery and in some instances might have resulted in elimination of impacted stocks. The information provided by the coded wire tag program also was critical to the success of some projects in the Sound Ecosystem Assessment program.

## **NEED FOR THE PROJECT**

### **A. Statement of Problem**

Wild pink salmon runs in Prince William Sound which were injured by the *Exxon Valdez* oil spill needed protection from overharvest during commercial fisheries. This was difficult to accomplish since these injured wild populations migrate through fishing areas with uninjured populations as well as large hatchery runs. It was not possible to simply close these fishing areas without severely affecting local and state economies. Inseason and postseason information on the mix of the various runs in fishing areas allowed fishery managers to direct fishing effort away from injured wild runs and to achieve desired spawning escapement goals.

Estimation of hatchery contributions from coded wire tag recoveries relies on several previously untestable assumptions. Among them are assertions pertaining to brood stock composition, tag loss, differential mortality, and straying. For some contribution estimates, particularly ones associated with the Northern district, there is suspicion that assumptions regarding tag loss and absence of wild fish in the brood pond are flawed. Cannery Creek contributions have been underestimated if these assumptions are invalid. Additionally, studies involving tag-placement and homing fidelity suggest coded wire tags can cause straying. This is a critical issue for a number of reasons. Current understanding of run timing and distribution of hatchery fish in mixed stock fisheries has been based on coded wire tag data. Without resolution of this problem, it will be impossible to determine if changes in run-timing detected from thermal mark recoveries are real or are a result of differences in methodology. With respect to estimation of hatchery contributions, a tag-induced stray fish no longer represents its original cohorts, and cannot be expanded normally. It is not known if tag placement affected the distribution of fish encountered in the commercial fisheries and cost recovery fisheries.

In the summer of 1997, coded wire tagged and otolith marked pink salmon will return to Prince William Sound, presenting a unique opportunity to examine the tenuous assumptions, and, if necessary, generate additional adjustments.

## **B. Rationale/Link to Restoration**

Coded wire tag application has been the tool of choice for uniquely marking hatchery pink salmon in Prince William Sound since 1986. This information has been used by fishery managers to direct fishing effort away from oil-damaged wild stocks.

Examination of the assumptions underlying estimation of hatchery contributions from coded wire tag recoveries will reveal sources of bias in the technique. The impact of any discovered bias will be discussed in the final report, and if necessary, additional adjustments to contribution estimates made. The resulting final report will paint a more complete picture of the coded wire tag estimation program, outlining its pitfalls and strengths, and so provide a more accurate assessment of estimated hatchery contributions.

## **C. Location**

This project was conducted in the Prince William Sound region. Pink salmon fry were marked at the three hatcheries operated by the Prince William Sound Aquaculture Corporation (Armin F. Koernig, Wally H. Noerenberg, and Cannery Creek) and the single hatchery operated by the Valdez Fisheries Development Association (Solomon Gulch). Sampling sites were dependent upon the disposition of the commercial and hatchery cost recovery harvests and occurred in various communities within Prince William Sound (i.e. Cordova, Valdez, and Whittier), and communities outside of Prince William Sound (Seward, Anchorage, Kenai and Kodiak). Some sampling was also done aboard processing vessels in Prince William Sound as well as at hatchery sites.

## **COMMUNITY INVOLVEMENT AND TRADITIONAL ECOLOGICAL KNOWLEDGE**

This program was cooperatively funded by both Prince William Sound Aquaculture Corporation and the Valdez Fisheries Development Association, the two private, nonprofit hatchery groups operating within Prince William Sound. These two groups are operated by a mix of individuals with ties to commercial, sport, personal use and subsistence fishing as well as by community representatives. Large scale tagging programs have been a cooperative effort between the Alaska Department of Fish and Game and these private, nonprofit aquaculture groups since the late 1980's.

Project plans and reports on results of the coded wire tag program have been reviewed by the Prince William Sound/Copper River Regional Planning Team as well as interested fishing industry groups. As part of the Trustee Council Natural Resource Damage Assessment and Restoration process, the coded wire tag recovery program has been subjected to extensive peer review and annual public review and comment. Results of the coded wire tag program were

presented at the March 1993 Oil Spill Symposium sponsored by the Trustee Council, the 1993 Pink and Chum Workshop, the annual spring meeting of the Prince William Sound Aquaculture Corporation Board of Directors, the 1994 Alaska Board of Fisheries meeting and the 1995 and 1996 Trustee Council Restoration Workshops.

The coded wire tag program has also been critical to the success of the integrated package of the Sound Ecosystem Assessment studies. The Sound Ecosystem Assessment program has roots in a broader plan developed by the Prince William Sound Fisheries Ecosystem Research Planning Group. Many Sound Ecosystem Assessment program projects depended upon information provided by coded wire tags.

The project employed local residents for data collection activities in fish processing plants located in Cordova, Valdez, Whittier, Seward, Anchorage, Kenai, and Kodiak, and at hatcheries in Prince William Sound. The project also employed residents of Juneau for tag extraction and decoding activities performed by the Alaska Department of Fish and Game Tag Laboratory. Permanent Alaska Department of Fish and Game Biologists stationed in Cordova and biometrics staff stationed in Anchorage completed data analyses and reports. Goods and services required by the project were obtained from vendors in the local communities where data was collected.

## **PROJECT DESIGN**

### **A. Objectives**

Trustee Council funds plus those contributed by the Alaska Department of Fish and Game, the Prince William Sound Aquaculture Corporation, and the Valdez Fisheries Development Association contributed to the completion of the following objectives for the 1995-1997 salmon seasons in Prince William Sound:

1. Using undecoded-tag data, provide timely inseason estimates of the temporal and spatial contributions of tagged hatchery stocks of pink salmon to Prince William Sound commercial and hatchery harvests.
2. Assess the properties of a new, faster, but potentially less reliable inseason estimator of contributions of tagged hatchery stocks, which is based upon undecoded tags and estimates of tender loads (catches).
3. Using decoded-tag data, provide hatchery-specific estimates of the temporal and spatial contributions of tagged hatchery stocks to the commercial and cost-recovery harvests in Prince William Sound.

4. Estimate marine survival rates for each uniquely coded hatchery release group of pink salmon.

Funding is requested in this close-out budget to achieve the following two objectives designed to test the validity of adjustment factor corrections.

5. Determine the incidence of stray fish and the rate of adipose-clipped fish without tags in the brood stocks of Prince William Sound hatcheries.
6. Determine the origin of adipose-clipped fish without tags recovered from Northern district catches.

Objective 5 will provide a test of the assumption that brood stocks contain only fish released from the hatchery and will identify the extent to which differential mortality and tag loss contribute to decreased tag rates in returning fish. Analyses of untagged clipped fish recovered from Northern district commercial catches (Objective 6) will complement investigations into tag loss at Cannery Creek. (\$10,000)

Recoveries of these fish will occur in the summer of 1997, with otolith readings and data analysis planned for FY98. All of the heads from adipose missing pink salmon recovered have in the past been sent to the CWT laboratory in Juneau for tag confirmation. Objectives 5 and 6 will be met by removing the otoliths from those salmon heads at the tag lab which did not contain tags and identifying them as to their origin (hatchery vs. hatchery vs. wild). This request is being made in the coded wire tagging project as it will provide information vital for a comprehensive final report.

## **B. Methods**

Personnel policy, purchasing practices, field camp operations, safety procedures, and project administration were in compliance the Alaska Department of Fish and Game Division of Commercial Fisheries Manual of Standard Operating Procedures (SOP). Data collection and estimation procedures were similar to those used in Natural Resource Damage Assessment Fish/Shellfish Study #3. These procedures have been thoroughly reviewed by the Natural Resource Damage Assessment peer review process and were approved by the Management Team.

### *Commercial and Cost-Recovery Harvests*

Recoveries were stratified by district, week, and processor. This stratification was chosen as a result of the findings of Peltz and Geiger (1990) who detected significant differences between the proportions of some tag codes among such strata. The differences indicate that processors tend

to receive catches from only certain parts of a district and is believed to be the result of traditional tendering patterns.

Recoveries of pink salmon tags from commercial and cost-recovery harvests were made as fish are pumped from tenders onto conveyor belts at land-based processors located in Cordova, Valdez, Seward, Anchorage, Whittier and aboard floating processors after each opening. Fish were sampled by technicians standing beside the belt. Each sampled fish was subjected to a visual and tactile examination for a missing adipose fin. It was never possible for an observer to census all fish from a tender during the unloading process. However, on occasion, holding tanks in processing plants contained fish from only one tender. In those instances it was possible for an observer standing on the processing line to get a census of an entire tender load which was previously sub sampled by technicians on the unloading conveyor. A Chi-square test of independence was used to compare the rate of occurrence of adipose fin clips in the census with that observed in the random sample from the load.

Data recorded for each tender included harvest type (i.e., commercial or cost-recovery catch), fishing district(s) from which the catch was taken, catch date, processor, and the number of fish examined. Catch data was obtained later from fish tickets.

Heads of adipose fin clipped fish were excised, identified with a uniquely numbered cinch strap, and bagged. These heads were then individually passed through a tag detector machine which produced an audible signal in the event that the head contains a coded wire tag. This procedure yielded numbers of undecoded tags in the sample. Heads were then frozen for subsequent shipment to the Alaska Department of Fish and Game coded wire tag Laboratory in Juneau (Tag Lab).

### *Brood Stock Harvests*

Tag shedding from release to return and differential mortality between tagged and untagged fish lead to discrepancies between marking rates at release and recovery. Hatchery brood stocks were scanned for tags in order to estimate adjustment factors which were used to account for the loss of tags from the population. Three assumptions inherent in the use of the brood stock for this purpose were: (1) the brood stock consisted solely of fish reared at the hatchery, (2) the tendency for a tagged fish to lose a tag or to die was similar for all fish marked at the same hatchery, and (3) for a specific tag code, the marking rate in the commercial fishery was the same as that in the brood stock.

It is believed that the first of these assumptions was violated at all facilities except at the W. H. Noerenberg hatchery (Sharr et. al. 1994f). Recovery of adult otoliths from this brood stock in 1997 will provide insight as to the validity of the assumption. A historical average adjustment factor calculated from the brood stock from the W. H. Noerenberg hatchery was considered an

appropriate quantity with which to adjust for tag loss and differential mortality for tags recovered from all facilities.

With respect to the second assumption, tagging practices vary little within a facility, and it was believed that the rate of tag loss and tag-induced mortality were similar for all fish tagged with a hatchery. There is suspicion, however, that tag loss rates for fish released from the Cannery Creek hatchery are much higher than those for fish released from the other Prince William Sound hatcheries. This can only be confirmed by examining otoliths sampled from the brood in 1997. All pink salmon heads have in the past been removed from all fish having a missing adipose fin and sent to the tag laboratory in Juneau for confirmation of tag presence. This year those heads that do not contain tags will have the otoliths removed and examined at the Juneau otolith laboratory to identify the origin of the fish as to which hatchery or if it is wild.

The third assumption relates to the possibility of tag-induced straying of hatchery fish away from the brood. Some histological evidence to this end was referenced in Sharr et al. (1994d). More direct preliminary evidence was discussed by Sharr et al. (1994f). The opportunity to study this relationship will only exist in the summer of 1997.

The adjustment factor for hatchery  $h$ ,  $a_h$ , was estimated as the ratio of sampled fish in the brood stock to the expanded number of fish based on tags found in the sample :

$$\hat{a}_h = \frac{s_h}{\sum_i \frac{x_i}{p_i}}$$

where,

$T$	=	number of tag codes released from hatchery $h$ ,
$p_i$	=	tagging rate at release for the $i$ th tag code (defined as number of tagged fish released with the $i$ th tag code divided by the total number of fish release group $i$ ),
$x_i$	=	number of tags of the $i$ th code found in $s_h$ and,
$s_h$	=	number of brood stock fish examined in hatchery $h$ .

The historical (1989-1996 for inseason, 1989-1997 for postseason) average W. H. Noerenberg adjustment factor was used to adjust contribution estimates (Equation 2) if it was shown that it was significantly greater than 1.0 at the 90% level.

While only the (historical) adjustment factor associated with the W. H. Noerenberg facility was used in any contribution estimation, brood stock samples were taken during hatchery egg-take

operations at each of the four Prince William Sound pink salmon hatcheries. Technicians, examined approximately 95% of the fish through visual and tactile means for missing adipose fins. The number of fish sampled were recorded and when adipose-clipped fish were found, the heads were excised and shipped on a weekly basis along with sample data to the Tag Lab.

#### *Tag Extraction, Tag Decoding, and Data Archiving*

During the fishing season all sampling data and heads from adipose-clipped fish were sent daily to the Alaska Department of Fish and Game Tag Lab. Data received at the Tag Lab were logged and tag recovery sampling forms were edited for accuracy and completeness. Samples which affected critical fisheries decisions were processed first. Tag lab staff located and removed tags from heads, decoded extracted tags, and entered tag code and sample data into a statewide database accessible to biologists in Cordova. Completed tag recovery data for prioritized samples were transmitted electronically to Cordova project personnel within 36 hours of the receipt of unprocessed data at the Tag Lab. In the following 12 hours Cordova project personnel integrated tag recovery and catch data from the Alaska Department of Fish and Game fish ticket reporting system to estimate hatchery and wild catch contributions. Contribution estimates were used by fisheries managers to implement the inseason management actions required.

Following the fishing season, processing of all lower priority tag recovery samples were completed by the Tag Lab. All tags recovered throughout the season were examined a second time to insure that they were properly decoded. All codes were validated with a master Pacific States Marine Fisheries Commission list of codes potentially present in Pacific coast fisheries. Fully edited tag code and sampling data from all samples collected during the season were forwarded to the Cordova office for final summarization and analyses. A complete historic database of coded wire tag information from Prince William Sound tagging and tag recovery programs is maintained by the Alaska Department of Fish and Game Tag Lab, the Pacific States Marine Fisheries Commission and, the Cordova office of the Alaska Department of Fish and Game. The Alaska Department of Fish and Game historic fish ticket catch database is maintained at the Alaska Department of Fish and Game Juneau headquarters office and in the Cordova area office. All tagging and recovery data and all fisheries harvest data are freely available from any of these sources.



## Postseason Hatchery Contributions and Survival Rates

The contribution of release group  $t$  to the sampled common property, cost-recovery, brood stock and special harvests, and escapement,  $C_t$ , were estimated as:

$$\hat{C}_t = \sum_{i=1}^L x_{it} \left( \frac{N_i \hat{a}_h}{s_i p_t} \right)$$

where,

- $x_{it}$  = number of group  $t$  tags recovered in  $i$ th stratum,
- $N_i$  = total number of fish in  $i$ th stratum,
- $s_i$  = number of fish sampled from  $i$ th stratum,
- $p_t$  = proportion of group  $t$  tagged,
- $\hat{a}_h$  = adjustment factor associated with hatchery  $h$ , and
- $L$  = number of recovery strata associated with common property, cost-recovery, brood stock, special harvests and escapement in which tag code  $t$  was found.

The contribution of release group  $t$  to unsampled strata,  $Cu_t$ , was estimated from contribution rates associated with strata which were sampled from the same district-week openings as the unsampled strata:

$$\hat{C}_t = \sum_{i=1}^U N_i * \left[ \frac{\sum_{j=1}^S \hat{C}_{tj}}{\sum_{j=1}^S N_j} \right]$$

where,

- $U$  = number of unsampled strata,
- $N_i$  = number of fish in  $i$ th unsampled stratum
- $S$  = number of strata sampled in the period in which the unsampled stratum resides,
- $\hat{C}_{tj}$  = contribution of release coded with tag  $t$  to the sampled stratum  $j$ , and
- $N_j$  = number of fish in  $j$ th sampled stratum.

When a district-week opening was not sampled at all (an infrequent occurrence), the catch from that opening was treated as unsampled catch of the subsequent opening in the same district. An estimate of the contribution of tag group  $t$  to the total Prince William Sound return was obtained through summation of contribution estimates for sampled and unsampled strata. An estimate of the total hatchery contribution to the Prince William Sound return was calculated through summation of contributions over all release groups. A variance approximation for  $C_t$ , derived by Clark and Bernard (1987) and simplified by Geiger (1988) was :

$$\hat{V}(\hat{C}_t) = \sum_{i=1}^L x_{it} \left[ \frac{N_i \hat{a}}{s_i p_t} \right] \left[ \frac{N_i \hat{a}}{s_i p_t} - 1 \right]$$

Assuming that covariance's between contributions of different release groups to a stratum can be ignored, summation of variance components over all tag codes provided an estimate of the variance of the total hatchery contribution. Inspection of the formula given by Clark and Bernard (1987) for the aforementioned covariance's shows them to be negligible for large  $N$  and  $s$ , and to be consistently negative, so that when ignored, conservative estimates of variance are obtained. Variances associated with unsampled strata are believed to be small (Sharr et al., 1994d).

The survival rate of the release group coded with tag  $t$  ( $S_t$ ), was estimated as:

$$\hat{S}_t = \frac{\hat{C}_t + \hat{C}_{ut}}{R_t}$$

where,

- $C_t$  = contribution of release coded with tag  $t$  to sampled strata,
- $C_{ut}$  = contribution of release group coded with tag  $t$  to unsampled strata,
- $R_t$  = total number of fish in release group coded with tag  $t$  released from hatchery.

Assuming the total release of fish associated with a tag code is known with negligible error, and that the cumulative variance contributions associated with the unsampled strata are small, a suitable variance estimate for  $S_i$  is given by:

$$\hat{V}(\hat{S}_i) = \frac{\sum_{i=1}^L x_{it} \left[ \frac{N_i \hat{a}}{s_i p_i} \right] \left[ \frac{N_i \hat{a}}{s_i p_i} - 1 \right]}{R_i^2}$$

### *Inseason Hatchery Contributions*

Inseason fisheries decisions which must be made on very short notice required rapid, real time analysis of coded wire tag data. Three inseason estimates of hatchery contributions of pink salmon were generated for each opening. The first and most timely estimate was calculated using knowledge of numbers of tags (undecoded) found in a sample taken from the catch and an estimate of that catch. The presence of tags in adipose clipped fish was discerned by passing their excised heads over a scanner identical to those used by the Tag Lab. The estimate of the catch aboard tenders was obtained from tender captains or processor operators. In the event that catch estimates could not be obtained, a simple unweighted average (over sampled tenders) proportion of hatchery fish in the catch was reported. Estimation using undecoded tags required that assumptions be made about expansion ( $1/p_i$ ) and adjustment ( $a$ ) factors (see Equation 2). For fishery openings in the western and northern portions of Prince William Sound, late-run returns from the Prince William Sound Aquaculture Corporation facilities were assumed to be the only hatchery contributors. For openings in the Southwestern district, an expansion factor which is a weighted average of all expansion factors associated with tags released at the A. F. Koernig, W. H. Noerenberg and Cannery Creek hatcheries was used. The weighting scheme depended upon historical contributions of hatcheries to the district in question. A similar weighting scheme for expansion factors was used for the Coghill and Northern districts and involved historical contributions associated with the Cannery Creek and W. H. Noerenberg hatcheries. For openings in the eastern part of Prince William Sound, returns to the Valdez Fisheries Development Association Solomon Gulch facility were assumed to be the only hatchery contributor. With respect to an appropriate expansion factor for these openings, the average of all factors associated with tags released from the Solomon Gulch facility was used. An average historical (1989-1997) adjustment factor associated with the W. H. Noerenberg facility was used for all inseason contribution estimates. These estimates could be made available at any stage of the unloading process, and only required that some sampling was conducted. The precision of the estimate was, of course, increased as more of the catch was

sampled. Such readily available, but less precise estimates played a significant role in those fishery management decisions that had to be made before the more precise estimates which required exact catch figures and larger sample sizes were available. Calculations of inseason contributions followed those used to generate post-season results (Equation 2). The second estimator was identical to the first, except that it was calculated only after sampling of an opening was completed and after exact tender loads were reported. The result was a less timely but more reliable estimate. The third estimator was less timely still because it relied on exact catch data and extracted and decoded tags. Use of code-specific expansion factors, however, provided hatchery-specific contribution estimates and mean a reduction in bias of the estimates resulting from use of average expansion factors.

### *Alternatives*

Estimation of stock- specific contributions to large commercial fisheries requires some sort of natural or man-induced mark which is characteristic of the stock or groups of stocks to be distinguished. Any mark used for estimates of stock specific catch contributions for inseason fisheries management must: (1) be naturally present in all or a fixed portion of the population or easy to apply permanently to a fixed portion of the population in the early life stages before stock mixing occurs, (2) be easy to distinguish in adult returns, (3) be present or can be applied to a large enough portion of the population such that significant numbers can be recovered among adult returns in a cost-effective manner for accurate and precise estimates of catch contributions, and (4) not affect survival or behavior of fish.

Until recently, coded wire tag technology has been the only man-induced mark available which meet most of the above criteria. Although this technology has provided the opportunity to distinguish hatchery and wild fish in commercial harvests, it is not without problems. The pink salmon tagging program in Prince William Sound is the largest of its kind in the world and is pushing the limit of the technology for both application and recovery. Application in very small fish such as pink salmon may affect survival, may not be permanent (tag loss), and tagging may affect behavior. Some methods exist and are used to adjust for tag loss from differential mortality and tag shedding. The effect of tag-induced straying, though thought to be small, is, however, difficult to accommodate. On the recovery side, large and expensive sampling programs must be implemented to ensure sufficient precision of contribution estimates.

Otolith marking methods meet all of the five criteria described above. Thermal marks have been thoroughly tested in all salmon species. They are permanent, are easily applied to every individual in a hatchery population and are less expensive to apply and recover relative to coded wire tags. Because they can be applied to every individual in the population, contribution estimates based on thermal marks will be more accurate and precise than those based on coded wire tags. Differential mortality of tagged fish will no longer be a problem. Because the mark is non intrusive, permanent tag loss through shedding and straying of tagged fish will also be eliminated. A large scale otolith marking program for Prince William Sound hatchery pink

salmon releases is underway. Recoveries of otolith marks from these releases will begin in 1997. This will be the only year of overlap between the two mark types. Starting in the summer of 1998 only otolith marked pink salmon will be returning to Prince William Sound.

Chemical marking of otoliths has not been tested in salmon to the same degree as thermal marking, but is widely used in other species. Chemical marking requires that young fish be fed or immersed in a chemical agent which leaves a recognizable band on otoliths or skeletal structures. Tetracycline is one widely used chemical which deposits a distinctive skeletal or otolith growth band which is florescent under ultraviolet light. Because it is retained in the tissues, Food and Drug Administration permits for its use in fish destined for human consumption fish were initially difficult to obtain but permitting is now done on a routine basis for many species. The method has promise for marking wild fish where heated water is not available for thermal marks.

To date no natural markers have been discovered in Prince William Sound pink salmon which allow researchers to distinguish hatchery stocks from all wild stocks. Genetic marks are a possibility but hatchery parent stocks in Prince William Sound originated from wild stocks in the area and are shared by more than one facility, and hence are probably not distinguishable.

### **C. Contracts**

This was a cooperative program funded by the Trustee Council, the Alaska Department of Fish and Game, the Prince William Sound Aquaculture Corporation, and the Valdez Fisheries Development Association. The Alaska Department of Fish and Game, Commercial Fisheries Management and Development Division ensured that 1) pink salmon catches were scanned for pink salmon with clipped adipose fins; 2) representative samples of heads from adipose-clipped pink salmon were collected and shipped to the Juneau Tag Laboratory; 3) information obtained from this project was adequately documented and cataloged, 4) biometrics review of methods and data analysis was obtained, and 5) reports documenting results were written. The Alaska Department of Fish and Game Tag Laboratory in Juneau extracted and decoded all coded-wire tags from samples of pink salmon heads sent from Prince William Sound. Funds from the Prince William Sound Aquaculture Corporation and the Valdez Fisheries Development Association for coded-wire tag recovery operations were conveyed to the Alaska Department of Fish and Game through cooperative agreements.

## **SCHEDULE**

### **A. Measurable Project Tasks for FY 98**

October 1997 - September 1998:	Analyze data collected in the summer of 1997 and write final report for project from years 1995 through 1997.
January 1998:	Attend Annual Restoration Workshop
September 30 1998:	Submit Final report for coded wire tag project

## **B. Project Milestones and Endpoints**

October - April 1998:	Analyze data for summer of 1997 return
April- September, 1998:	Write Final report for coded wire tagging project

## **C. Completion Date**

The Trustee Council had originally approved two years of overlap between the coded-wire tag and otolith marking programs, with funding for final data analysis and report writing being made available in FY 99. This multi-year project will now be completed a year earlier, in FY 98, because coded wire tags were not applied to emergent pink salmon in the spring of 1997. There are two reasons for this action. First the local private non-profit hatchery organizations maintained that they could not afford to apply tags. Second, highly visible thermal marks were applied to brood year 1996 pink salmon, and the private non-profit organizations argued that application of coded wire tags was unnecessary.. The 1997 season will now be the last year to recover tags and FY98 funds will be used for final data analysis and report writing. Otolith thermal marking will thereafter be used to provide stock assessment. The quality and readability of the otolith marks applied in the winters of 1995 and 1996 were high, and the technique should be functional in 1998.

## **PUBLICATIONS AND REPORTS**

The final project report will be submitted by September 30, of 1998.

## **PROFESSIONAL CONFERENCES**

Project results should be presented at either the Alaska Chapter of the American Fisheries Society Annual meeting or the biennial Pink and Chum Salmon Workshop or both. The next occurrence of these meetings after the data are collected and analyzed will be the fall of 1998 and spring of 1999.

## **NORMAL AGENCY MANAGEMENT**

The *Exxon Valdez* Trustee Council has played a major role in the development of pink salmon stock identification in Prince William Sound. The Trustee Council provided support for the coded wire tag program during the damage assessment phase because the project provided essential information for evaluating injury to salmon stocks as well as invaluable data used by managers to direct harvests away from damaged wild stocks. The program has been jointly

funded during the Restoration phase with the Trustee Council contributing nearly half of the funds, the remainder being contributed by the Prince William Sound Aquaculture Corporation, the Valdez Fisheries Development Association and the Alaska Department of Fish and Game. Although the coded wire tag program provided data of a sort previously unavailable to managers, shortcomings in the technique became apparent as the project evolved. The most significant shortcoming was related to tag expansions and was associated with the inability to mark all fish in the population. It was at this time that large-scale thermal mass marking emerged as a promising new methodology, and funds were sought from the Trustee Council to implement the program in Prince William Sound. A timeline and budget had been formulated for the near-future development of the Prince William Sound stock identification program. It consisted of continued development of the thermal mass marking program, with two years of overlap with the coded wire tag program. For reasons stated above that timeline was changed so that only one year of overlap exists. Budgets have been adjusted in both the coded wire tag program and the otolith thermal marking program to reflect this change (Table 1). By FY 2000, program funding for the otolith thermal marking and recovery will be the responsibility of the Prince William Sound Aquaculture Corporation, the Valdez Fisheries Development Association and the Alaska Department of Fish and Game.

Table 1. Budgets for otolith marking and coded wire tagging programs for stock-identification of pink salmon in Prince William Sound (thousands of dollars).

	FY96	FY97	FY98	FY99	FY2000
<b>CWT<sub>a</sub> program</b>					
	Recover BY 94 Tag BY 95	Recover BY 95	Reports		
Trustee Council	248.6	273.8	126.6	0.0	0.0
ADF&G <sub>b</sub>	81.6	56.8	89.0	0.0	0.0
PWSAC/VFDA <sub>d</sub>	277.6	262.0	0.0	0.0	0.0
Total	607.8	592.6	215.6	0.0	0.0
<b>Otolith program</b>					
	Mark BY 95 Sampling Experiments	Mark BY 96 Recover BY 95	Mark BY 97 Recover BY 96	Mark BY 98 Recover BY 97	Mark BY 99 Recover BY 98
Trustee Council	93.2	120.1	141.1	182.9	0.0
ADF&G	0.0	57.5	56.3	138.3	158.0
PWSAC/VFDA	0.0	64.1	113.0	155.6	155.0
Total	93.2	241.7	310.4	476.8	313.0
<b>Total Program</b>					
Trustee Council	341.8	393.9	267.7	182.9	0.0
ADF&G	81.6	114.3	145.3	138.3	158.0
PWSAC/VFDA	277.6	326.1	113.0	155.6	155.0
Grand Total	701.0	834.3	526.0	476.8	313.0

a Coded wire tag

c Prince William Sound Aquaculture Corporation

b Alaska Department of Fish and Game

d Valdez Fisheries Development Association



## COORDINATION AND INTEGRATION OF RESTORATION EFFORT

The foundations for this project were firmly established in joint feasibility studies which were conducted by the Alaska Department of Fish and Game and non-profit aquaculture associations in Prince William Sound beginning in 1986 and extending through 1988. Results of these studies have been summarized by Peltz and Miller (1990), Peltz and Geiger (1990), and Geiger and Sharr (1990). During the damage assessment process large scale tagging and recovery projects were instituted and perfected by Natural Resources Damage Assessment Fish/Shellfish Study #3. Damage assessment funds were expended for tagging hatchery releases of pink salmon in 1989 and 1990 and wild populations of pink salmon in 1990 and 1991 (Fish/Shellfish Study #3). Tag recovery efforts for wild and hatchery pink salmon were funded by damage assessment funds in 1989, 1990, and 1991 (Fish/Shellfish Study #3) and by restoration funds in 1992 and 1993 (Restoration Studies 60A and 93067). Results of damage assessment and restoration coded wire tag studies have been reported by Sharr et al. (1994d, 1994e and 1994f, 1995) and Riffe (1996). Following the loss of funds for further tagging of hatchery stocks of pink salmon in 1990, the private non-profit aquaculture groups in Prince William Sound continued to tag pink salmon releases at their own expense. Tags applied to pink fry from the four pink salmon hatcheries in Prince William Sound in 1996 must be recovered to provide comparative data for the otolith mark recoveries beginning in 1997.

The pink salmon coded wire tag recovery program has complimented several other projects since 1989. Improved escapement estimates for Prince William Sound pink salmon from Natural Resource Damage Assessment Fish/Shellfish Study 1 and restoration Study 60B were used in conjunction with catch contribution estimates from the coded wire tag recovery projects to adjust fishery exploitation rates and achieve wild stock escapements. Growth and survival estimates from Fish/Shellfish Study #4 could not have been obtained without Fish/Shellfish Study #3, which provided tagged fish of known origin and release timing. The pink salmon coded wire tag recovery program was also integrated with several other salmon restoration projects being conducted in Prince William Sound in 1996. It complemented the Sound Ecosystem Assessment program, the multi-disciplinary program designed to develop an understanding of the mechanisms regulating ecosystem function in Prince William Sound. The Sound Ecosystem Assessment program is focused on interactions of pink salmon and herring with other components of the Prince William Sound ecosystem. Marked pink salmon provide a valuable tool for examining interactions between wild and hatchery salmon during the early marine period. The salmon growth component of the Sound Ecosystem Assessment program uses marked pink salmon to evaluate habitat overlap between wild and hatchery salmon, to examine the size composition of wild and hatchery salmon in mixed schools, and to estimate juvenile salmon mortality during the time of ocean residence. The salmon predation component of the Sound Ecosystem Assessment program uses marked pink salmon to determine whether predators select wild or hatchery salmon.

## **EXPLANATION OF CHANGES IN CONTINUING PROJECTS**

A major change has occurred in the project design and schedule described in the approved FY97 Detailed Project Description. Pink salmon fry were not tagged in the spring of 1997 because of a financial crisis within the private non-profit hatcheries in Prince William Sound. As a result, the hatchery produced pink salmon adult return of 1998 will not possess coded wire tags, and the only method available to separate stocks will be by otolith thermal marks. It is, therefore, imperative that any comparative studies be conducted in 1997. Both the coded wire tag program and otolith program budgets have been adjusted to reflect changes elicited by not applying coded wire tags to hatchery pink salmon released in 1997. A large reduction in the coded wire tag budget has occurred and a smaller yet significant increase in the otolith budget was required to meet the program needs.

## **PROPOSED PRINCIPAL INVESTIGATOR**

Timothy L. Joyce  
Alaska Department of Fish and Game  
Commercial Fisheries Management and Development Division  
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Cordova, Alaska 99574-669  
(907) 424-3214 (phone)  
(907) 424-3235 (FAX)  
timj@fishgame.state.ak.us (E-mail)

## **PERSONNEL**

The Principal Investigator (PI) for the project will be a permanent full-time Fisheries Biologist III (FB III) for the Alaska Department of Fish and Game. The PI will be responsible for writing project operational plans, administering project budgets, quality control of data collection, supervising data analyses and, co-authoring final reports. A Fisheries Biologist II (FB II) will, maintain data quality, assist in data analyses, and co-author final reports. Many of the FB II day to day activities will be shifted to the otolith project. The FB II will be assisted in the otolith project by one permanent seasonal Fisheries Biologist I (FB I). The FB I will be in charge of supervising day to day sampling activities in Cordova and do first reads on otoliths that arrive in Cordova.

A Biometrician I from the Alaska Department of Fish and Game Commercial Fisheries and Development Division Region II office in Anchorage will provide biometrics support for the

project. The Biometrician I will assist in experimental design, inseason and post season data analyses, and report writing.

The Biometrician I will travel to Cordova several times during the year to assist with inseason data analyses and final data analyses and report writing.

#### TIMOTHY L. JOYCE - Fisheries Biologist III Principal Investigator

Mr. Joyce has a Bachelor of Science in Fisheries Science from Oregon State University (1973). Mr. Joyce was appointed to the Fisheries Biologist III position in July of 1995. Prior to this appointment he worked for the State of Alaska as a hatchery manager at Kitoi Bay which was the largest multi-species salmon production facility run by the state. He did some of the initial half-length coded wire tag work on emergent pink salmon fry from 1982 through 1987. He co-authored an article titled "Retention Rates of Half-Length Coded Wire Tags Implanted in Emergent Pink Salmon" published in the American Fisheries Society Symposium 7:253-258, 1990. He has over 17 years experience in salmon hatchery production in Alaska working with all five species of Pacific salmon, but primarily with pink salmon. Prior to his position as the hatchery manager at Kitoi Bay, Mr. Joyce worked in Sand Point, Alaska as a high school teacher instructing in Aquaculture, fish culture and biology. He was responsible for a small demonstration hatchery run by the school district with Johnson O'Malley funds where students had hands on training of salmon culture using pink and coho salmon. Mr. Joyce also has extensive experience in warm water fish culture gained while in Africa working as a Peace Corps volunteer at a UN development project under the FAO.

#### RENATA RIFFE - Fisheries Biologist II Research Biologist

Ms. Riffe has a Master of Science in Statistics from Colorado State University (1994), a Master of Science in Fisheries Management from the University of Alaska, Fairbanks (1987), and a Bachelor of Science in Fishery Biology from Colorado State University (1981). Since October 1994 Ms. Riffe has worked on the coded wire tag project as an FB II Research Biologist in the capacity of Assistant Project Leader. Prior to her current position, (from June 1991 - October 1994), she was employed as a biologist with the Alaska Department of Fish and Game, Sport Fish Division in Fairbanks, Alaska, and assisted in projects concerning abundance estimation and population evaluation of pike, grayling, humpback whitefish, least cisco, rainbow trout, burbot, chum salmon, and king salmon. From May 1982 - January 1991, she worked as a technician with the Alaska Department of Fish and Game, Commercial Fisheries Management and Development Division in Juneau, Alaska. Her primary duties involved sampling commercial salmon fisheries and salmon escapements, with some report writing. She also developed discriminant function models for stock separation of Lynn Canal sockeye salmon, by scale pattern analysis, developed a computer model which simulated migratory timing of salmon

escapements, and evaluated truncated escapement counts. She has authored reports for the Alaska Department of Fish and Game on estimates of abundance and survival rates of round whitefish, compilation of age and length data for rainbow trout in southwest Alaska, and migratory timing of salmon in the Situk River, Alaska.

#### DAVID EVANS - Biometrician I

Mr. Evans has a Bachelor of Science in soil science from the University of Nottingham (U.K.), a Master of Science and a Doctor of Philosophy degree in soil science from the University of Guelph (Ontario, Canada), and a Master of Science in statistics from Oregon State University. David has worked with the Alaska Department of Fish and Game since October, 1991. His primary responsibility has been analysis of coded-wire-tag data from Prince William Sound. He has designed the statistical procedures and computer spread sheets used for inseason analysis of tag recovery data, has overseen most of the post season data analyses and has co-authored interim and final reports for the 1991 Fish /Shellfish Study #3, the 1992 Restoration Study 60C, and the 1993 Restoration study 93067.

#### LITERATURE CITED

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# 1998 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1997 - September 30, 1998

Budget Category:	Authorized FY 1997	Proposed FY 1998					
Personnel		\$100.2					
Travel		\$0.0					
Contractual		\$10.6					
Commodities		\$0.0					
Equipment		\$0.0					
Subtotal	\$0.0	\$110.8					
General Administration		\$15.8					
Project Total	\$0.0	\$126.6					
Full-time Equivalents (FTE)		1.8					
Dollar amounts are shown in thousands of dollars.							
Other Resources							
Comments:							
Close-out of CWT project Trustee Portion = \$126.6 K							
Additional Funding Sources ADF&G Portion = \$89.0K PNP Portion = \$0 K							
CLOSE - OUT FUNDING Included in close-out funding in FFY98 is \$10,000 to determine origin of adipose clipped pink salmon that do not contain a tag by reading the otolith marks inorder to test assumptions used in the adjustment factor for stock contribution. Cannery Creek contributions may have been greatly underestimated.							

1998

Project Number: 98186  
Project Title: Coded Wire Tag Recoveries from Pink Salmon,  
PWS  
Agency: AK Dept. of Fish and Game

FORM 3A  
TRUSTEE  
AGENCY  
SUMMARY

**1998 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET**

October 1, 1997 - September 30, 1998

<b>Personnel Costs:</b>		GS/Range/ Step	Months Budgeted	Monthly Costs	Overtime	Proposed FY 1998
Name	Position Description					
Tim Joyce	Fishery Biologist III	18L	3.0	7.0	0.0	21.0
Renate Riffe	Fishery Biologist II	16D	3.0	5.2	0.0	15.6
David Evans	Biometrician I	17F	5.0	6.0	0.0	30.0
Melanie Guerrero	F & W Tech. III	11A	1.0	3.8	0.0	3.8
Tag Lab technicians	Juneau technicians	9A	9.3	3.2	0.0	29.8
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
<b>Subtotal</b>			21.3	25.2	0.0	
<b>Personnel Total</b>						<b>\$100.2</b>
<b>Travel Costs:</b>		Ticket Price	Round Trips	Total Days	Daily Per Diem	Proposed FY 1998
Description						
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
<b>Travel Total</b>						<b>\$0.0</b>

**1998**

Prepared: 2 of 4

Project Number: 98186  
 Project Title: Coded Wire Tag Recoveries from Pink Salmon,  
 PWS  
 Agency: AK Dept. of Fish and Game

FORM 3B  
 Personnel  
 & Travel  
 DETAIL

4/14/97



**1998 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET**  
October 1, 1997 - September 30, 1998

<b>Contractual Costs:</b>		Proposed
Description		FY 1998
Office costs		3.6
Publication costs		7.0
When a non-trustee organization is used, the form 4A is required.		
<b>Contractual Total</b>		<b>\$10.6</b>
<b>Commodities Costs:</b>		Proposed
Description		FY 1998
<b>Commodities Total</b>		<b>\$0.0</b>

**1998**

Project Number: 98186  
Project Title: Coded Wire Tag Recoveries from Pink Salmon,  
PWS  
Agency: AK Dept. of Fish and Game

FORM 3B  
Contractual &  
Commodities  
DETAIL

**1998 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET**  
October 1, 1997 - September 30, 1998

[illegible]

## 1998

Project Number: 98186  
Project Title: Coded Wire Tag Recoveries from Pink Salmon,  
PWS  
Agency: AK Dept. of Fish and Game

FORM 3B  
Equipment  
DETAIL

Prepared: 4 of 4

414/97



## **Otolith Thermal Mass Marking of Hatchery Reared Pink Salmon in Prince William Sound**

Project Number:	98188
Restoration Category:	General Restoration
Proposer:	Alaska Department of Fish and Game
Lead Trustee Agency:	Alaska Department of Fish and Game
Cooperating agencies:	Prince William Sound Aquaculture Corporation Valdez Fisheries Development Association
Alaska SeaLife Center:	
Duration:	Fourth year, five-year project
Cost FY 98:	\$141,100
Cost FY 99:	\$182,900 (Close-out)
Cost FY 00:	\$0
Cost FY 01:	\$0
Cost FY 02:	\$0
Cost FY 03:	\$0
Geographic Area:	Prince William Sound
Injured Resource/Service:	Pink Salmon

**RECEIVED**  
'APR 15 1997  
**EXXON VALDEZ OIL SPILL  
TRUSTEE COUNCIL**

### **ABSTRACT**

This project develops otolith mass marking as a technology for identification of hatchery pink salmon returning to Prince William Sound. The otoliths of all pink salmon reared at Prince William Sound hatcheries will be thermally marked in the fall of 1998. A blind test will be conducted to determine the ability of otolith readers to successfully determine the origin of randomly selected otoliths. During the 1998 commercial fishery, approximately 100 otoliths will be processed from each fishery opening to estimate stock composition. A Bayesian approach will be used in the estimation of postseason contribution estimates, with a dynamic sample size allocation scheme being used to maximize sampling efficiency

## INTRODUCTION

Each year approximately 500 million wild pink salmon fry emerge from the streams of Prince William Sound and migrate seaward. Annual adult runs of wild pink salmon to Prince William Sound have averaged 10 million salmon over the last two decades. The large migrations of fry and subsequent adult runs of pink salmon play major roles in the Prince William Sound ecosystem. Both juveniles and adults are important sources of food for many fish, birds, and mammals. Adults returning from the high seas also convey needed nutrients and minerals from the marine ecosystem to estuaries, freshwater streams, and terrestrial ecosystems. Wild pink salmon also play a major role in the economy of Prince William Sound because of their contribution to commercial, sport, and subsistence fisheries in the area.

Up to 75% of the pink salmon spawning habitat in Prince William Sound occurs in intertidal areas. In the spring of 1989 oil from the *T/V Exxon Valdez* spill was deposited in intertidal portions of many western Prince William Sound streams. Pink salmon embryos and fry rearing in these intertidal areas appear to have been adversely affected by the oil. Sharr et al. (1994a and 1994b) observed salmon embryo mortalities which were 67%, 51%, 96%, and 80% higher in oiled streams than in nearby comparable unoiled streams in 1989, 1990, 1991, and 1992, respectively. Wiedmer (1992) also observed a high incidence of deformities and elevated levels of cytochrome P-450 among fry in oiled streams in 1989, and Willette (1993) reported reduced growth and survival of pink salmon fry and juveniles which reared in oiled marine waters of Prince William Sound in 1989. Mortality differences between oiled and unoiled streams in 1989 and 1990 were confined to intertidal spawning areas and may be attributed to direct lethal effects of oil. Large differences observed across all tide zones in 1991 and 1992 may be the consequence of damage to germ cells of the adults which originated from the 1989 and 1990 brood years when egg and larval exposures to intertidal oil were greatest. A consequence of this genetic damage may be persistent functional sterility and reduced returns per spawner for populations from oiled streams.

Prince William Sound pink salmon returns originating from brood years following the *Exxon Valdez* oil spill have been aberrant. Returns of wild and hatchery pink salmon in 1991 were only slightly below the midpoint of the preseason forecast but arrived late and had very compressed run timing. The salmon were also small and in advanced stages of sexual maturity long before reaching their natal streams. As a result, the salmon were of little commercial value. Returns of pink salmon in 1992 and 1993 were far fewer than expected. The 1992 return of wild pink salmon was the fourth smallest even year return in the last 30 years, and the hatchery return was less than one third of the expected. The 1993 return of wild pink salmon was the third smallest in the last 30 years, and the hatchery return was less than one fifth of the expected. The returns in 1994, 1995 and 1996 were much larger than those in 1992 and 1993, but were skewed to the eastern portion of Prince William Sound.

Although hatchery pink salmon production in Prince William Sound began in the 1970's, returns from maximum authorized levels of fry production did not occur until the late 1980's and early 1990's, which coincided with the *Exxon Valdez* oil spill era. The migratory timing of wild salmon populations injured by the *Exxon Valdez* oil spill is similar to that of hatchery stocks, so wild and hatchery salmon are exploited together in commercial, sport, and subsistence fisheries.

To sustain production from wild populations, managers must insure that adequate numbers of wild salmon from all portions of the wild run escape fisheries and enter streams to spawn. To achieve this goal, mixed stock fisheries must be managed to achieve exploitation rates appropriate for less productive wild populations. To accomplish these rates, managers must be able to distinguish wild from hatchery salmon and estimate their relative spatial and temporal abundance in different fishing areas. The otolith-marking program is designed to accomplish this task in an efficient and comprehensive manner.

In 1995 and 1996, pink salmon otoliths were thermally marked at all Prince William Sound hatcheries under projects R96188 and R97188, respectively. These studies showed that marked otoliths were highly readable and that proposed catch sampling methodologies were appropriate. In 1997, the first estimates of hatchery contributions based on recovery of marked otoliths will be made. These estimates will be compared to those based on coded wire tag recoveries, which will allow an evaluation of some important assumptions inherent in that program to be conducted. Work planned for FY98 includes continuing the otolith marking program, testing readability of marks, and estimating contributions of hatchery salmon marked in 1996.

## **NEED FOR THE PROJECT**

### **A. Statement of the Problem**

Coded wire tags have been the tool of choice for applying unique marks to hatchery pink salmon in Prince William Sound. The method has been used extensively to estimate hatchery and wild stock contributions to commercial harvests and has also been used to study straying. Trustee Council projects F/S3, R60C, R93067, R94320b, R95320b, R96186 and R97186 have all incorporated this technology to estimate contributions of wild and hatchery pink salmon returns to Prince William Sound since the *Exxon Valdez* oil spill. Despite its usefulness, there are disadvantages to coded wire tag technology. Approximately 1 million coded wire tags must be applied to pink salmon fry each year to obtain catch contribution estimates for returning adults. Tagging and recovery are both very labor intensive, and the number of tags applied and recovered are sometimes inadequate for the levels of accuracy and precision desired. Coded wire tags are intrusive, can be shed, and may affect subsequent survival. Tag loss through shedding and differential mortality of tagged individuals affects subsequent estimates of adult returns based on tag recoveries. Finally, recent evidence suggests that poor placement of coded wire tags may cause salmon to stray (Habicht (*personal communication*; found in Appendix D of Seeb et al. 1995)). Therefore, a new technology with fewer disadvantages is needed to effectively separate stocks of pink salmon returning to Prince William Sound.

### **B. Rationale/Link to Restoration**

Because of the cost and problems associated with coded wire tag technology, other alternatives for marking larger portions of populations with less expensive and intrusive methods must be investigated. This project will develop otolith mass marking as an inseason and postseason stock separation tool for pink salmon. By marking otoliths of all of salmon in a population, sample sizes in the recovery phase may be much smaller without affecting accuracy and precision of contribution estimates. The nonintrusive, permanent nature of otolith marking eliminates

concerns over mark shedding and marking effects on survival and behavior, all of which may be important sources of error in coded wire tag estimates. Numerous studies have documented the successful induction of predetermined ring codes on fish otoliths by manipulation of water temperature during embryonic stages (Bergstedt et al. 1990, Brothers E.B. 1990, Munk and Smoker 1990, Volk et al. 1990). Each of these studies has provided information regarding the magnitude of temperature differences and the duration of temperature cycles needed to produce otolith rings. Recognizing the need to develop mass marking technology for pink salmon in Prince William Sound, the Alaska Department of Fish and Game and Prince William Sound Aquaculture Corporation reviewed the feasibility of otolith thermal marking at Prince William Sound hatcheries and otolith recovery in commercial fisheries (Geiger et al. 1994). An otolith marking and recovery program conducted during 1993 in Southeast Alaska (Hagen et al., 1995) developed an inseason otolith sampling and mass processing protocol which appeared to be suitable for Prince William Sound. Additional work is needed to fully develop and evaluate otolith thermal marking technology as an inseason fisheries management tool for Prince William Sound pink salmon.

In order to provide information that can be used to regulate fisheries during the season, recovered otoliths need to be examined and decoded within 24 hours. Establishing an otolith dissecting and reading laboratory in Cordova in cooperation with the central processing laboratory in Juneau will provide management biologists with timely information on stock composition for inseason management decisions. A single, coordinated database accessible to both laboratories will be developed to ensure proper quality control and archival of data.

### **C. Location**

The study will be undertaken in Prince William Sound. One benefit of the project will be a sustainable local fishery, which will result in more stable local economies (fishermen, cannery employees, local retailers, tourism, etc.). With improved protection of wild pink salmon stocks in Prince William Sound, one of the most important driving forces of that ecosystem will be assured, resulting in maintenance of local ecological diversity and enhancement of desire by the general public to visit this area.

## **COMMUNITY INVOLVEMENT AND TRADITIONAL ECOLOGICAL KNOWLEDGE**

This project was developed through three months of ecosystem research planning by the Prince William Sound Fisheries Ecosystem Research Planning Group as part of the Sound Ecosystem Assessment program. The Prince William Sound Fisheries Ecosystem Research Planning Group conducted public meetings each week in the fall of 1993. Scientists from the University of Alaska, University of Maryland, Prince William Sound Science Center, Prince William Sound Aquaculture Corporation, the Alaska Department of Fish and Game, and US Forest Service participated in the planning process. The resulting ecosystem research plan was reviewed by scientists from the United States and Canada at a public workshop held in Cordova, Alaska in early December 1993. The methods and results of this project will continue to be reviewed by various scientists within the Program Management component of the Sound Ecosystem Assessment program.

This project is partially sponsored by the Prince William Sound Aquaculture Corporation, the regional private, non-profit (PNP) aquaculture association for Prince William Sound, and the Valdez Fisheries Development Association, a small PNP operation. Development of production level marking programs, such as the Prince William Sound coded wire tagging program, have been a cooperative effort between the Alaska Department of Fish and Game and Prince William Sound area private non-profit (PNP) aquaculture associations since the early 1980's. PNP's, operated by a broad constituency of commercial, sport, personal use, and subsistence fishers and community representatives, reviewed coded wire tag project plans and results annually before approving subsequent funding. Operational plans and results of marking projects are also reviewed periodically by the Prince William Sound/CR Regional Planning Team as well as interested fishing industry groups. As part of the Trustee Council NRDA and Restoration process the coded wire tag marking and recovery project has been subjected to extensive peer review and annual public review and comment. Results of coded wire tag projects were presented at the March 1993 Oil Spill Symposium sponsored by the Trustee Council, the 1993 Pink and Chum Workshop, the annual Spring meeting of the Prince William Sound Aquaculture Corporation board of directors in 1993, and the Alaska Board of Fisheries in 1994. The Prince William Sound Aquaculture Corporation and the Valdez Fisheries Development Association board of directors as well as the Prince William Sound/CR Regional Planning Team have endorsed development of otolith thermal mass marking of hatchery salmon in Prince William Sound as an alternative to coded wire tagging.

## **PROJECT DESIGN**

### **A. Objectives**

During FY 98 the following four objectives will be achieved:

1. Application of otolith thermal marks to all pink salmon embryos incubating in the A. F. Koernig (AFK), W. H. Noerenberg (WHN), Cannery Creek (CC), and Solomon Gulch (SG) hatcheries.
2. Evaluation of the quality of otolith thermal marks applied to pink salmon embryos at AFK, WHN, CC, and SG hatcheries, including collection of voucher samples needed when examining otoliths from returning adults.
3. Estimation of stock composition of commercial catches and hatchery brood stock collections of pink salmon using otolith thermal marks.
4. Evaluation of the quality of stock estimation procedures.

### **B. Methods**

#### *Objective 1*



All pink salmon embryos will receive a hatchery specific base mark on their otoliths after reaching the eyed stage. At the W. H. Noerenberg and A. F. Koernig hatcheries, some pink salmon will receive an accessory mark after hatching (alevin stage) to identify rearing strategies. To avoid mortalities in marking alevins, both facilities have installed the necessary equipment to avoid gas-bubble disease.

A unique thermal base mark will be used for each hatchery in Prince William Sound (Table 1). A consistent, unique base mark will simplify both application and recovery of marks. Thermal base marks will be applied in the zone of the otolith corresponding to the period between eyed-embryo and hatch stages.. This period occurs between September and December and has an average length of 35 days. Approximately 22 days will be required to apply thermal base marks at each hatchery. Although the length of hot and cold water events may change to reflect fish culture concerns at the time of marking, the assigned mark pattern will remain the same. Accessory marks applied to some of the production at the W. H. Noerenberg and A. F. Koernig hatcheries will be composed of three thermal rings. A minimum of six days will be required to apply this mark.

Table 1. Otolith base marks assigned to Prince William Sound pink salmon hatcheries. The thermal schedule is the actual temperature regime where "H" refers to relatively hot water and "C" refers to relatively cold water. The difference between "H" and "C" at each hatchery is 4C °. The number preceding each "H" and "C" is the number of hours embryos are reared at that temperature level. Terms in parentheses denote the number of repetitions needed to form the desired ring pattern.

Hatchery	Thermal Schedule	Ring pattern
A. F. KOERNIG	(4X)24H:24C	IIII
Accessory	(3X)24H:24C	III
CANNERY CREEK	(3X)24H:24C,(1X)72H:36C,2(X)24H:24C	III III
W.H. NOERENBERG	(8X)24H:24C	IIIIIIII
Accessory	(3X)24H:24C	III
SOLOMON GULCH	(6X)24H:24C	IIIIII

Evaluation of thermal marks will be made in two steps.

1) Characterization of applied marks.

Characterization of actual banding patterns achieved by the marking process will be accomplished through use of otolith 'voucher sets' representing all manifestations of a mark at a given hatchery. Each voucher set will contain samples of otoliths from each incubator in the hatchery. Voucher sets will be formed for all four hatcheries producing pink salmon, and will be available to otolith readers working with 1998 commercial catch samples. Initial otolith voucher set preparation and assessment will be conducted at the ADF&G Otolith Laboratory in Juneau. This work will consist of extraction and grinding procedures to expose internal structures, followed by examination of ring patterns under a compound microscope. Quality control and data archival will also be the responsibility of the Otolith Laboratory in Juneau.

2) Determination of the success of the marking process.

The success of the marking process will be evaluated based on: 1) photomicrographs of voucher otoliths; 2) agreement among technicians in assessing otolith banding patterns for juvenile pink salmon sampled during early marine survival studies; 3) a blind test using otoliths from local wild and hatchery fry, in which otolith readability is assessed as the probability that the origin of a randomly selected otolith can be correctly determined by Otolith Laboratory technicians.

The first evaluation method will consist of a visual examination of the base marks applied at the hatcheries. Otolith laboratory staff will rate marks as excellent, good or poor. A poor rating would indicate that problems are likely to occur in identifying those marks in mixed stock samples. The second method will use agreement between readers examining otoliths from juveniles captured on their seaward migration. Examining otoliths from mixed stock samples should more accurately reflect the mix of patterns that may be expected in returning adults. The third method will use a blind test to determine the probability that otoliths from known populations of hatchery and wild fry can be correctly identified. This method will be similar to the blind test used in 1996 but will be done on a smaller scale. The following information will be derived from the readings: 1) overall readability of otoliths from each facility, including an associated confidence interval; 2) an identification matrix, in which misclassifications to specific populations are recorded.

*Objective 3*

The composition of pink salmon catches in 1998 will be estimated using recovered thermal marks. Technicians will sample tender boats delivering pink salmon to processors using a sampling methodology developed during the 1996 season. Systematic samples will be taken from each tender delivering salmon from an opening. Once the total catch from the fishery opening is known, otoliths will be sampled from each tender collection in proportion to the load aboard the tenders so that 100 otoliths can ultimately be chosen from that opening. Otoliths

collected but not used in the 100 salmon sample will be stored for possible processing after the season during the dynamic sampling allocation phase (Geiger 1994). Sampling 100 otoliths from each fishery opening should yield estimates of the proportion of hatchery salmon in the catch which are approximately within  $\pm 10\%$  of the true proportion 95% of the time. Actual precision will probably be greater than since it is likely that the proportion of hatchery salmon will deviate from the worse case scenario of a 50% catch contribution. The precision of the total season estimate of the contribution of hatchery stocks to the harvest will depend upon the actual number of pink salmon harvested in each opening. An analysis of harvests from previous years indicates that the precision of this estimate will be approximately  $\pm 2\%$  of the true proportion 95% of the time, and  $\pm 2\%$  of the true proportion greater than 95% of the time when the proportion of hatchery salmon in some or all of the fishery openings deviates from 50%. Inseason processing of otoliths will be conducted at the Alaska Department of Fish and Game Area Office Laboratory in Cordova, and quality control of the process will be provided by personnel from the Juneau Otolith Laboratory. Postseason analysis using dynamic sampling allocation will follow the methods of Geiger (1994).

Otoliths will also be recovered from adult pink salmon used during egg-take operations at all Prince William Sound hatcheries. These samples will be used to estimate the composition of the brood stock collections. This information will be used to test the assumption, used in the coded wire tag program, that only the hatchery brood stock which does not contain wild salmon is the at the W. H. Noerenberg facility.

#### *Objective 4*

The quality and utility of the stock estimation procedure will be evaluated in terms of accuracy and precision of in- and postseason estimates, quickness in providing estimates to fishery managers during the season, and accuracy of otolith identification.

### **C. Cooperating Agencies, Contracts, and Other Agency Assistance**

The Alaska Department of Fish and Game Commercial Fisheries Management and Development Division will ensure that (1) information obtained from this project is adequately documented and catalogued, and (2) biometrics review of project methods and data analyses is obtained. The Alaska Department of Fish and Game Otolith Laboratory will assist in processing all otolith samples collected during this project. Since this project is a cooperative study conducted jointly by the Alaska Department of Fish and Game, the Prince William Sound Aquaculture Corporation, and the Valdez Fisheries Development Association, contractual service agreements will be needed for application and recovery of thermal marks from each private non-profit agency.

## **SCHEDULE**

### **A. Measurable Project Tasks for FY 98**

This project will be conducted over one pink salmon life cycle for both odd- and even-brood year populations. Embryos will be otolith marked in the fall of 1997 and 1998. Pink salmon from the 1996 and 1997 brood years will return to Prince William Sound as adults in the summers of 1998 and 1999. The following tasks will be accomplished in FY 98:

October to December:	Apply thermal marks to BY 97 embryos at four pink salmon hatcheries
November-January:	Develop FY 99 DPD and FY98 annual report
February-March:	Collect samples from incubators to evaluate thermal mark quality
March-June:	Process and evaluate otoliths
April 15:	Submit annual project report for FY 1997
June-September:	Collect otoliths, process otoliths, analyze data, make recommendations
April 1999:	Submit annual project report for FY 1998

#### **B. Project Milestones and Endpoints**

The following milestones and endpoints will be achieved from FY 98 onward.

December 1997:	Objective 1 - Apply thermal marks to brood year 1997 embryos
June 1998:	Objective 2 - Evaluate thermal mark quality for brood year 1997
February 1998:	Objective 3 - Estimate harvest stock composition for brood year 1995
February 1998:	Objective 4 - Evaluate quality of estimation procedure for brood year 1995
February 1999:	Objective 3 - Estimate harvest stock composition for brood year 1996
February 1999:	Objective 4 - Evaluate quality of estimation procedure for brood year 1996

#### **C Completion Date**

All objectives of this multi-year project are expected to be met by FY99. At that time, support for a fully developed inseason stock separation program will likely be shared by the Alaska Department of Fish and Game and the private sector.

#### **PUBLICATIONS AND REPORTS**

An annual project report will be submitted by April 15 of each year.

#### **PROFESSIONAL CONFERENCES**

Project results may be presented at technical meetings such as Alaska Chapter of the American Fisheries Society Annual meeting or the biennial Pink and Chum Salmon Workshop. The next occurrence of these particular meetings after data are collected and analyzed will be the fall of 1998 and spring of 1999.

## NORMAL AGENCY MANAGEMENT

The *Exxon Valdez* Trustee Council has played a major role in the development of pink salmon stock identification in Prince William Sound. The Trustee Council provided support for the coded wire tag program during the damage assessment phase because the project provided essential information for evaluating injury to salmon stocks as well as for directing harvest effort away from damaged wild stocks. The program has been jointly funded during the Restoration phase with the Trustee Council contributing nearly half of the funds and the remainder being contributed by the Prince William Sound Aquaculture Corporation, the Valdez Fisheries Development Association and the Alaska Department of Fish and Game. Although the coded wire tag program provided similar data, various disadvantage of the technique became apparent as the project evolved. The most significant relates to adjustment factors used to expand coded wire tag recoveries into estimates of actual numbers of pink salmon. This will not be a problem with otolith thermal marking since all salmon in produced by each hatchery will be marked. Development of a such a large-scale thermal mass marking program has only been possible using funds obtained from the Trustee Council to supplement other funds available for Prince William Sound. A timeline and budget was formulated for the Prince William Sound stock identification program which consisted of continued development of the thermal mass marking program along with two years of overlap with the existing coded wire tag program (Table 2). Unfortunately, the precipitous drop in the price of pink and chum salmon created extreme financial hardships for private sector cooperators, and they were unwilling to mark fry with coded wire tags as well as apply otolith marks in 1997. Double marked adults with both coded wire tags and otolith marks will, therefore, only be available for study during 1997. The close relationship of the two programs and the intermixing of personnel will result in an increased operating cost for the otolith project in FY98 and a decreased cost for the coded wire tagging project. By FY 2000, program funding will be the responsibility of the Prince William Sound Aquaculture Corporation, the Valdez Fisheries Development Association and the Alaska Department of Fish and Game.

Table 2      Budgets for otolith marking and coded wire tagging programs for stock-  
identification of pink salmon in Prince William Sound (thousands of dollars).

	FY96	FY97	FY98	FY99	FY2000
<b>CWT<sub>a</sub> program</b>					
	Recover BY 94 Tag BY 95	Recover BY 95	Reports		
Trustee Council	248.6	273.8	126.6	0.0	0.0
ADF&G <sub>b</sub>	81.6	56.8	89.0	0.0	0.0
PWSAC/VFDA <sub>d</sub>	277.6	262.0	0.0	0.0	0.0
Total	607.8	592.6	215.6	0.0	0.0
<b>Otolith program</b>					
	Mark BY 95 Sampling Experiments	Mark BY 96 Recover BY 95	Mark BY 97 Recover BY 96	Mark BY 98 Recover BY 97	Mark BY 99 Recover BY 98
Trustee Council	93.2	120.1	141.1	182.9	0.0
ADF&G	0.0	57.5	56.3	138.3	158.0
PWSAC/VFDA	0.0	64.1	113.0	155.6	155.0
Total	93.2	241.7	310.4	476.8	313.0
<b>Total Program</b>					
Trustee Council	341.8	393.9	267.7	182.9	0.0
ADF&G	81.6	114.3	145.3	138.3	158.0
PWSAC/VFDA	277.6	326.1	113.0	155.6	155.0
Grand Total	701.0	834.3	526.0	476.8	313.0

a Coded wire tag

b Alaska Department of Fish and Game

c Prince William Sound Aquaculture Corporation

d Valdez Fisheries Development Association

## **COORDINATION AND INTEGRATION OF RESEARCH EFFORT**

The Otolith Mass Marking Project (98188) is integrated with several other salmon restoration projects in Prince William Sound. This project will complement the Sound Ecosystem Assessment program (Project 98320). The Sound Ecosystem Assessment program is a multi-disciplinary program designed to develop an understanding of the mechanisms regulating ecosystem function in Prince William Sound. The Sound Ecosystem Assessment program is focused on interactions of pink salmon and herring with other components of the Prince William Sound ecosystem. Otolith marked salmon will provide a valuable tool for examining interactions between wild and hatchery salmon during the early marine period. The salmon growth component of the Sound Ecosystem Assessment program will utilize otolith marked juvenile pink salmon to (1) evaluate habitat overlap between wild and hatchery salmon, (2) examine size composition of wild and hatchery salmon in mixed schools, and (3) to estimate juvenile salmon mortality within Prince William Sound and the Gulf of Alaska. The salmon predation component of the Sound Ecosystem Assessment program will utilize otolith marked juvenile salmon to determine if predators select wild or hatchery salmon.

The Prince William Sound Aquaculture Corporation and the Valdez Fisheries Development Association will use thermal mass marking to place unique marks on the otoliths of all pink salmon fry released from their facilities in brood year 1997. Both private non-profit organizations will provide personnel to recover otoliths from pink salmon delivered to processors in Prince William Sound. The Alaska Department of Fish and Game Otolith Laboratory in Cordova will process otoliths recovered from the common property and cost recovery fisheries and broodstocks in order to make inseason estimates of contributions. The Alaska Department of Fish and Game Otolith Laboratory in Juneau will provide quality control and technical support to the Cordova Laboratory and Prince William Sound hatcheries.

## **EXPLANATION OF CHANGES IN CONTINUING PROJECTS**

The code wire tag program will end one year sooner than originally planned since the private non-profit hatcheries did not apply tags to emerging pink salmon fry in 1997. As a result, the otolith program will be the only tool available for stock separation in the summer of 1998. The coded wire tag and otolith marking projects are closely related, and in the interest of efficiency, several personnel work on both projects. Since staff essential to the otolith program are partially funded through the coded wire tag program, an increase in the requested otolith budget is necessary in order to maintain the program in a functional form.

## **PROPOSED PRINCIPAL INVESTIGATOR**

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## **PERSONNEL**

The Principal Investigator (PI) for the project will be a permanent full-time Fisheries Biologist III (FB III) working for the Alaska Department of Fish and Game. The PI will be responsible for writing project operational plans, administering project budgets, quality control of data collection, supervising data analyses and, co-authoring final reports. A Fisheries Biologist II (FB II) will supervise day to day project operations, maintain data quality, assist in data analyses, and co-author final reports. The FB II will be assisted by one permanent seasonal Fisheries Biologist I (FB I). The FB I will be in charge of supervising day to day sampling activities in Cordova and will assist the PL in supervising sampling at other ports, on floating processors, and at hatcheries. Non-permanent Fish and Wildlife Technician III's (FWT III) will be stationed in Cordova and Valdez and will assist the FB I as crew leaders. The crews in each port will be non-permanent FWT II's. Under the supervision of the FB I, the FWT III's will conduct daily data logging, editing and archiving activities in Cordova and Valdez.

A Biometrician I from the Alaska Department of Fish and Game Commercial Fisheries and Development Division Region II office in Anchorage will provide biometrics support for the project. The Biometrician I will assist in experimental design, inseason and post season data analyses, and report writing.

The PI, FB II or, FB I will maintain daily phone contact with project technicians stationed in ports other than Cordova or Valdez and at several remote hatchery locations. Copies of data forms from these sites will be faxed to Cordova daily and otoliths from sampled salmon will be shipped once or twice weekly to Cordova via scheduled commercial flights or via chartered aircraft depending upon which is available. The PL, APL, or project Fisheries Biologist I will make routine supervisory visits to each sampling port via chartered or commercial aircraft at least twice monthly for sampling quality control inspections, data collections, and industry contacts. The Biometrician I will travel to Cordova several times during the season to assist with inseason data analyses and occasionally after the season to assist with final data analyses and report writing.



TIMOTHY L. JOYCE - Fisheries Biologist III, Principal Investigator  
Alaska Department of Fish and Game; Commercial Fisheries Management and Development  
P. O. Box 669, Cordova, Alaska 99574  
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#### EMPLOYMENT:

Mr. Joyce was appointed to the Fisheries Biologist III position in July of 1995. Prior to this appointment he worked for the State of Alaska as a hatchery manager at Kitoi Bay which was the largest multi-species salmon production facility run by the state. He did some of the initial half-length coded wire tagging work on emergent pink salmon fry from 1982 through 1987. He co-authored an article titled "Retention Rates of Half-Length Coded Wire Tags Implanted in Emergent Pink Salmon" published in 1990 in the American Fisheries Society Symposium 7:253- 258. He has over 17 years experience in salmon hatchery production in Alaska working with all five species of Pacific salmon, but primarily with pink salmon. Prior to his position as the hatchery manager at Kitoi Bay, Mr. Joyce worked in Sand Point, Alaska as a high school teacher instructing in Aquaculture, fish culture and biology. He was responsible for a small demonstration hatchery run by the school district with Johnson O'Malley funds where students had hands on training of salmon culture using pink and coho salmon. Mr. Joyce also has extensive experience in warm water fish culture gained while in Africa working as a Peace Corps volunteer at a UN development project under the FAO.

#### OTHER EXPERIENCE:

Commercial herring spotter pilot, 1985 -1994  
Research aid, Oak Creek Laboratory, Corvallis, OR. 11/71 - 6/73  
Construction, 1964 - 1971

#### EDUCATION:

1973: Bachelor of Science , Fisheries Science, Oregon State University.

RENATA RIFFE - Fisheries Biologist II, Research Biologist  
Alaska Department of Fish and Game; Commercial Fisheries Management and Development  
P. O. Box 669, Cordova, Alaska 99574  
Ph. (907) 424-3212

#### EMPLOYMENT:

Since October 1994 Ms. Riffe has worked on the coded wire tag project as an FB II Research Biologist in the capacity of Assistant Project Leader. Prior to her current position, (from June 1991 - October 1994), she was employed as a biologist with the Alaska Department of Fish and Game, Sport Fish Division in Fairbanks, Alaska, and assisted in projects concerning abundance estimation and population evaluation of pike, grayling, humpback whitefish, least cisco, rainbow trout, burbot, chum salmon, and king salmon. From May 1982 - January 1991, she worked as a technician with the Alaska Department of Fish and Game, Commercial Fisheries Management and Development Division in Juneau, Alaska. Her primary duties involved sampling commercial salmon fisheries and salmon escapements, with some report writing. She also

developed discriminant function models for stock separation of Lynn Canal sockeye salmon, by scale pattern analysis, developed a computer model which simulated migratory timing of salmon escapements, and evaluated truncated escapement counts. She has authored reports for the Alaska Department of Fish and Game on estimates of abundance and survival rates of round whitefish, compilation of age and length data for rainbow trout in southwest Alaska, and migratory timing of salmon in the Situk River, Alaska.

**EDUCATION:**

1994: Master of Science , Statistics, Colorado State University.

1987: Master of Science, Fisheries Management, University of Alaska, Fairbanks.

1981: Bachelor of Science, Fish Biology, Colorado State University.

VACANT - Fisheries Biologist I

**EMPLOYMENT:**

**EDUCATION:**

**PETER HAGEN - Cooperating Investigator (Otolith Laboratory Director)**

Department of Fish and Game, Commercial Fisheries Management and Development Division

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**EMPLOYMENT:**

August 1991 - Present: Director of the Alaska Department of Fish and Game Otolith Aging Laboratory. This laboratory was established to extract information from calcified tissues to aid in the management of the State of Alaska's fisheries resources. Responsibilities include implementing a program for mass marking hatchery reared salmon by imposing patterns on their otolith microstructure through temperature manipulation in the egg and alevin stages. The laboratory recovers the patterns from the otoliths of adult salmon to determine the proportion of hatchery salmon in mixed stock fisheries. The laboratory is also charged with aging groundfish using otoliths and other hard structures. The aging information is used to determine the status of stocks and is incorporated into age-structured population models. Responsibilities include developing research and project operation plans, instigating new cooperative studies, supervising laboratory personnel, budget management, coordinating activities with outside agencies, and other Fish and Game divisions.

September 1987 - 1991: Co-principal investigator of a joint Alaska Sea Grant - International Pacific Halibut Commission project investigating annuli and microstructure patterns in otoliths of Pacific halibut. This project is being used to complete a Ph.D. in Fisheries. It involves innovative use of image processing, x-ray microscopy, and statistical methodology to describe the process of otolith growth and quantify pattern variation. The research includes an analysis of the historical collection of otoliths maintained by the International Pacific Halibut Commission. The otolith collection provides a unique opportunity to develop a long-term record of otolith growth. This research is directed toward determining which quantifiable features of the otolith (both patterns and elemental composition) can be used to investigate mechanisms responsible

for long-term changes in population structure. Published results include identifying a long-term response of juvenile halibut growth to temperature changes. Additional work investigates the potential for identifying sub-stocks of halibut through trace elements incorporate into the otolith microstructure.

#### OTHER EXPERIENCE:

Fisheries Biologist, National Marine Fisheries Service, Auke Bay 6/86 - 9/87.  
Research Fellowship, International Pacific Halibut Commission, Seattle WA. 1/84 - 5/86  
Fisheries Consultant, 5/83- 9/84,  
Commercial Fisherman, 4/83  
Fisheries Biologist, International Pacific Halibut Commission, Seattle WA. 6/80 - 9/82

#### EDUCATION:

1994 Doctor of Philosophy (Candidate) Fisheries, University of Alaska, Fairbanks  
1986 Master of Science, Fisheries, University of Alaska, Juneau  
1981 Bachelor of Science, Fisheries Science, University of Washington

KRISTEN M. MUNK - Cooperating Investigator (Otolith Laboratory Biologist)  
Alaska Dept. of Fish & Game, Commercial Fisheries Management and Development Division  
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#### EMPLOYMENT:

Fisheries Biologist responsible for developing mass-processing techniques for recovery of otolith thermal marks, coordinating and conducting age analyses of groundfish structures, and supervising production of otolith processing and age structure information in the Alaska Department of Fish and Game-CFMD Otolith Lab.

1976 - present: Field experience includes gillnet test fishing in Lower Cook Inlet; commercial catch sampling of ship- and land-based processors in Cook Inlet and Prince William Sound; remote-site escapement sampling along the Kenai Peninsula, Prince William Sound and Southeastern Alaska; abundance surveys of sablefish using long lines; creel censusing of sport fishers in the Susitna drainage and Juneau marine waters; assisting in crab index surveys; flying aerial surveys for salmon; assisting in installation, operation, and maintenance of MTS and Bendix sonar; collecting habitat assessment data on numerous Juneau area creeks and rivers; enforcing fishing regulations; supervising field crews; conducting data analyses, under supervision, of commercial catch age data; aging scales; collecting fish using various trapping methods. Hatchery and weir experience includes installing, operating, and maintaining weirs; collecting data and keeping records; tagging and fin-clipping juvenile salmon; supervising tagging and weir crews; sampling tissue used for genetic stock identification studies; spawning salmon for aquaculture operations; transporting and placing eggs in incubators; monitoring incubation of salmon eggs; administering prophylactics; monitoring, maintaining, and releasing pen-reared salmon. Lab experience includes recovering and identifying aquatic insects and salmon fry stomach contents; preparing and aging otoliths, shark spines, and lingcod fin spines; designing,

implementing, coordinating otolith sampling programs; sampling , preparing and analyzing otolith samples; reporting on thermal mass marking projects and technology.

**EDUCATION:**

1989 Bachelor of Science, Zoology, University of Hawaii

**DAVID EVANS - Cooperating Investigator (Biometrician)**

Department of Fish and Game, Commercial Fisheries Management and Development Division

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Anchorage, Alaska 99518

**EMPLOYMENT:**

October, 1991 - present: Biometrician I with the Alaska Department of Fish and Game. Primary responsibility has been analysis of coded-wire-tag data from Prince William Sound. Design of the statistical procedures and computer spread sheets used for inseason analysis of tag recovery data. Oversight of most of the post season data analyses and co-author of interim and final reports for the 1991 NRDA F/S Study #3, the 1992 Restoration Study 60C, and 1993 Restoration studies 93137 and 93184.

**EDUCATION:**

1991 Master of Science, Statistics, Oregon State University

1988 Doctor of Philosophy, Soil Science, University of Guelph (Ontario, Canada)

1984 Master of Science, Soil Science, University of Guelph (Ontario, Canada)

1981 Bachelor of Science, Soil Science, University of Nottingham (U.K.)

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**1998 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET**  
October 1, 1997 - September 30, 1998

Budget Category:	Authorized FY 1997	Proposed FY 1998					
Personnel		\$110.0					
Travel		\$2.4					
Contractual		\$2.1					
Commodities		\$6.5					
Equipment		\$3.5					
Subtotal	\$0.0	\$124.5					
General Administration		\$16.6	Estimated FY 1999	Estimated FY 2000	Estimated FY 2001	Estimated FY 2002	
Project Total	\$0.0	\$141.1	\$182.9	\$0.0	\$0.0	\$0.0	
Full-time Equivalents (FTE)		1.8					
Dollar amounts are shown in thousands of dollars.							
Other Resources							
Comments:							
OTOLITH RECOVERY OPERATION WITHOUT CWT RECOVERY SINCE NO CWT'S APPLIED IN SPRING OF 1997							
Trustee Portion: = \$141.1 K							
Additional Funding Sources							
ADF&G Portion: = \$56.3 K							
PNP Portion: = \$113.0 K							

**1998**

Project Number: 98188  
Project Title: Otolith Mass Marking of Hatchery Pink Salmon in PWS  
Agency: AK Dept. of Fish and Game

FORM 3A  
TRUSTEE  
AGENCY  
SUMMARY



# 1998 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1997 - September 30, 1998

Personnel Costs:		GS/Range/	Months	Monthly		Proposed
Name	Position Description	Step	Budgeted	Costs	Overtime	FY 1998
Tim Joyce	Fishery Biologist III	18L	3.0	7.0	0.0	21.0
Renate Riffe	Fishery Biologist II	16D	3.0	5.2	0.0	15.6
Vacant Cordova	Fishery Biologist I	14A	4.0	4.2	2.7	19.5
Vacant Cordova	Fish & Wildlife Tech. III	11A	4.0	3.8	2.3	17.5
Otolith lab personnel	FB I	16D	2.0	5.3	0.0	10.6
(Juneau)	F&W Tech.'s	11A	2.0	3.9	0.0	7.8
David Evans	Biometrician I	17F	3.0	6.0	0.0	18.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
Subtotal			21.0	35.4	5.0	
Personnel Total						\$110.0
Travel Costs:		Ticket	Round	Total	Daily	Proposed
Description		Price	Trips	Days	Per Diem	FY 1998
Anchorage- Cordova; Biometrics support on catch sampling		0.2	1	3	0.1	0.5
Cordova - Anchorage; PI attendance at EVOS workshop		0.2	1	3	0.1	0.5
Cordova - Juneau; Otolith lab training and Quality Control		0.4	2	6	0.1	1.4
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
Travel Total						\$2.4

1998

Project Number: 98188  
 Project Title: Otolith Mass Marking of Hatchery Pink Salmon in PWS  
 Agency: AK Dept. of Fish and Game

FORM 3B  
 Personnel  
 & Travel  
 DETAIL

Prepared: 2 of 4

4/14/97

**1998 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET**

October 1, 1997 - September 30, 1998

<b>Contractual Costs:</b>		Proposed
Description		FY 1998
Air charter for training at hatcheries and otolith recovery crews to floating processors. 6 hrs @ \$250/hr		1.5
Freight for consumables		0.6
When a non-trustee organization is used, the form 4A is required.		
<b>Contractual Total</b>		<b>\$2.1</b>
<b>Commodities Costs:</b>		Proposed
Description		FY 1998
Grinding paper 4 boxes of 100 sheets @ \$75/box		0.3
petrographic slides 10 pkgs of 10 gross @ \$290/pkg		2.9
Petrographic slide boxes 50 @ \$16/ea.		0.8
Thermal plastic glue 5 pkgs of 6 bars @ \$67/pkg		0.3
96 cell flat bottom trays & lid 2 cases of 50 @ \$144		0.3
gloves, knives, labels, rubber bands, etc.		0.4
Office costs		1.5
<b>Commodities Total</b>		<b>\$6.5</b>

**1998**

Project Number: 98188  
 Project Title: Otolith Mass Marking of Hatchery Pink Salmon in PWS  
 Agency: AK Dept. of Fish and Game

FORM 3B  
 Contractual &  
 Commodities  
 DETAIL

**1998 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET**

October 1, 1997 - September 30, 1998

New Equipment Purchases:		Number of Units	Unit Price	Proposed FY 1998
Description				
	Bar Code Scanner & printer	1	3.5	3.5
				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 a <b>New Equipment Total</b>				\$3.5
Existing Equipment Usage:		Number of Units	Inventory Agency	
Description				
	Boiler module (Temp. inc.21 deg. F. at 200gpm)	1	VFDA	
	Boiler module (Temp. inc.21 deg. F. at 200gpm)	2	PWSAC	
	Boiler module (Temp. inc.21 deg. F. at 200gpm)	1	ADF&G	
	MZ6 Dissecting microscope	2	ADF&G	
	DMLS Binocular microscope	2	ADF&G	
	Labapol-5 grinders	2	ADF&G	

**1998**

Project Number: 98188

Project Title: Otolith Mass Marking of Hatchery Pink Salmon in PWS

Agency: AK Dept. of Fish and Game

FORM 3B  
Equipment  
DETAIL

Prepared: 4 of 4

4/14/97



## Construction of a Linkage Map for the Pink Salmon Genome

Project Number: 98190  
Restoration Category: Research  
Proposer: Fred W. Allendorf  
University of Montana  
Lead Trustee Agency: ADFG  
Alaska SeaLife Center: yes  
Duration: 3rd year, 5-year project  
Cost FY 98: \$211,100  
Cost FY 99: \$187,000  
Cost FY 00: \$187,000  
Geographic Area: Prince William Sound  
Injured Resource: Pink salmon

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EXXON VALDEZ OIL SPILL  
TRUSTEE COUNCIL

### ABSTRACT

We propose to complete our construction of a detailed genetic linkage map for pink salmon by analyzing the genetic transmission of several hundred DNA polymorphisms. The ability to map the genetic location of oil induced lesions will allow the thorough identification, description, and understanding of oil induced genetic damage. This research will also aid other recovery efforts with pink salmon, including estimation of straying rates, description of stock structure, and testing if marine survival has a genetic basis. We will complete the linkage map ahead of schedule in this the third year of our support. We propose to begin efforts to achieve Objectives 5 & 6 of this project using the Alaska SeaLife Center facilities.

## INTRODUCTION

We propose to complete a genetic linkage map for the pink salmon (*Oncorhynchus gorbuscha*) genome. Such a map will provide the necessary platform for identifying genetic damage in pink salmon inhabiting oiled streams following the March 1989 *Exxon Valdez* oil spill (EVOS). A detailed genetic map will also aid other recovery efforts with pink salmon, including estimation of straying rates, description of stock structure, and testing if marine survival has a genetic basis.

This project began in FY 96. However, we did not receive authorization to proceed until half-way through FY 96 (March 1996). Nevertheless, we have made substantial progress toward completing project objectives. This more rapid progress is the result of using two different and more efficient techniques for detecting genetic variation that were not included in the original proposal (PINEs and AFLPs; see description in **Methods**). We propose to complete the linkage map ahead of schedule by the end of FY 98, rather than by December 1999 as stated in our original DPD. In addition, we propose to take advantage of the early availability of the Alaska SeaLife Center and begin experiments that apply the linkage map to an understanding of the fundamental population biology and genetics of pink salmon. This work will provide important support for the existing project *Genetic structure of Prince William Sound pink salmon* (196).

We continue to pursue additional funding of this research through other sources. Aspects of this work are currently being supported by a grant from the National Science Foundation (Conservation and genetics of Pacific salmonids; DEB-9300135). In addition, Fred Allendorf is a member of a committee on Salmonid Genome Mapping for the U.S. Department of Aquaculture (USDA) chaired by Dr. Gary Thorgaard, Washington State University. Additional funding from the USDA will be requested after this committee has developed a cooperative plan among the projects that are involved.

Genetic linkage maps have provided the necessary information for understanding genetic variation in species since the rediscovery of Mendel's principles early in this century. A genetic map plays a similar role for a geneticist that a geographical map plays for the explorer of new territories. For many years, genetic maps could only be constructed in a very few model species that were suitable for extensive genetic manipulation (e.g., *Drosophila* and mice). Recent advances in molecular genetics now make it possible to uncover enough genetic markers to construct a detailed genetic linkage map in almost any species (Postlethwait et al. 1994).

This work will have significance for work with pink salmon under the project Oil-Related Embryo Mortalities (Restoration Study 191A). The objective of that project is to identify germline mutations in pink salmon exposed to oil. Genetic damage induced by oil may either be small changes in nucleotide sequence (microlesions) or large-scale changes in chromosome structure (macrolesions). A detailed genetic map for pink salmon will be invaluable for interpreting the results of Restoration Study 191A in several ways. First, it will be possible by following the inheritance of any DNA lesions to determine if they are micro- or macro-lesions. Second, these lesions can be mapped to determine if they are randomly spread throughout the genome or if they occur at mutational "hot spots" that are susceptible to oil induced damage.

The construction of a detailed linkage map will also serve as a basis for understanding genetic aspects of pink salmon restoration and supplementation. This work will be performed on both odd- and even-year pink salmon because of the known genetic differences between these fish. In addition, the outbreeding depression found in hybrids suggests that there are chromosomal differences between odd- and even-year fish (Gharrett and Smoker 1991). In addition, Phillips and Kapuscinski (1988) have described a translocation chromosomal polymorphism in odd-year pink salmon. Understanding the inheritance of allelic variation and linkage relationships is especially important and valuable in pink salmon, and other salmonid fishes, because of their polyploid ancestry (Allendorf and Thorgaard 1984).

## **NEED FOR THE PROJECT**

### **A. Statement of Problem**

Elevated embryo mortalities were detected in populations of pink salmon inhabiting oiled streams following the March 1989 *Exxon Valdez* oil spill (EVOS). These increased rates of mortality persisted through the 1993 field season, three generations after the oil spill, suggesting that genetic damage may have occurred as a result of exposure to oil during early developmental life-stages. The consequences of the putative genetic damage include impaired physiological function of individuals and reduced reproductive capacity of pink salmon populations.

The aggregate of evidence from the field studies and incubation experiment suggests that embryos exposed to oil in 1989 and 1990 accumulated deleterious mutations in the germline (reviewed in Detailed Project Description of Project 95191A). This hypothesis of genetic damage is consistent with previous field observations and laboratory experiments on the effects of crude oil on early life stages of fish. Long term intra-gravel oil exposures (7-8 months) to freshly fertilized eggs provide embryos sufficient time to accumulate polynuclear aromatic hydrocarbons (PAH's) from very low aqueous concentrations of crude oil. PAH's are abundant in crude oil and are potent clastogens (i.e. capable of breaking chromosomes).

Mironov (1969) observed reduced survival of fish embryos and larvae exposed to very low aqueous doses (1 ul oil/l seawater) of oil. Longwell (1977) reported genetic damage in pelagic embryos affected by the ArgoMerchant oil spill. Moles et al. (1987) confirmed that pink salmon embryos take up PAH's and demonstrated that the uptake was much greater in an intertidal environment than in strictly freshwater conditions. Biggs et al. (1991) found greater numbers of chromosome aberrations in larval herring which incubated in oiled areas than in non-oiled areas. It is likely that the same type of damage may have occurred in pink salmon, and this damage could have affected the germline of exposed individuals (Malkin 1994).

### **B. Rationale**

The recovery objective for pink salmon is healthy and productive populations that exist at prespill levels or levels in unoiled areas. An indication of recovery is when egg mortality in

oiled areas match prespill or levels in unoiled areas. A genetic map will be essential for detecting and understanding causes of reduced egg and embryo survival in oiled areas.

The genetic damage caused by exposure to oil may persist longer in populations of pink salmon than in other vertebrates because of the tetraploid nature of the salmonid genome. Salmonid fishes went through a tetraploid event some 25 million years ago that duplicated their entire genome (Allendorf and Thorgaard 1984). The extra genes in pink salmon may mask the effects of mutational damage caused by recessive deleterious alleles. The effects of these deleterious mutations may be uncovered in subsequent generations.

This fundamental genetic information will be of great assistance for three of the four Components of the Pink Salmon Restoration Program:

*Toxic Effect of Oil on Pink Salmon:* genetic mapping is essential for identifying genetic lesions induced by exposure to oil.

*Stock Separation and Management:* the genetic markers identified in the course of this study will provide greatly increased power and resolution to identify stocks of pink salmon on a very fine scale. In addition, determining the adaptive significance of these genetic markers will provide important information in interpreting the significance of genetic differentiation among pink salmon population samples.

*Supplementation:* the genetic markers will also be of great value in genetically identifying fish from supplementation programs and detecting their ecological and genetic interactions with wild fish.

Information gained from this study will provide resource managers with insight into the magnitude and persistence of damages sustained by wild pink salmon due to EVOS. Efforts to restore damaged pink salmon populations depend upon the ability of fishery managers to identify sources of reduced survival and to monitor their persistence. The potential of long term oil exposures to cause genetic damage needs to be understood so that spawning escapement goals can be adjusted if necessary. In addition, verification of the genetic hypothesis will provide the first evidence that the germline of fish exposed to chronic or acute sources of oil pollution can be affected.

Our results may have relevance for other fish species as well (e.g., Pacific herring, *Clupea pallasii*). Comparative gene mapping has shown that the linkage groups in a wide variety of vertebrates have been conserved (O'Brien et al. 1993). If there are certain loci in pink salmon that are mutational "hotspots" for oil induced damage, it would be possible to look for similar hotspots in Pacific herring or other fish species (e.g., rockfish, *Sebastes*).



## C. Location

Gametes for the inheritance studies have been collected from Prince William Sound in collaboration with the project Oil-Related Embryo Mortalities (Restoration Study \191A). Embryo incubation has taken place at the Genetics Lab facilities of ADFG. The initial laboratory phases of the project are being done at the University of Montana.

We propose to use the Alaska SeaLife Center Research Facilities at Seward for rearing fish and laboratory analyses. This facility will greatly strengthen genetic investigations with pink salmon by allowing multigenerational studies and testing for effects of specific genotypes on phenotypes of importance (marine survival, run timing, etc.). We anticipate that much of the laboratory analysis will be performed at this facility when it is available.

## COMMUNITY INVOLVEMENT

This is a specialized project that will not benefit directly from the knowledge of local/traditional people. We will hire local residents when possible for assistance (e.g., maintaining of fish). In addition, as a professional educator in a university I am very committed to educational efforts. I have contacted Ted Cooney in regard to giving a paper at the Arctic Division of the AAAS in Valdez in 24-27 September 1997, and I am interested in suggestions of other opportunities for informational meetings in the communities of Prince William Sound, including the Alaska SeaLife Center in Seward, and articles in the Trustee Council newsletter.

## PROJECT DESIGN

### A. Objectives

Our primary objective is to construct a detailed genetic linkage map for pink salmon by analyzing the genetic transmission of several hundred DNA polymorphisms. Pink salmon have 26 pairs of chromosomes ( $2N=52$ ; Allendorf and Thorgaard 1984), and, therefore, should have a total of 27 linkage-groups (LG's): 25 autosomes, an X-chromosome, and a Y-chromosome. We plan to map enough variable markers so that a new marker, such as a putative lesion identified in Restoration Study \191A, can be assigned with high probability to one of the 27 LG's. It is impossible to know how many markers this will require because we do not know the total length of the pink salmon linkage map. The linkage map of the zebrafish (*Danio rerio*) has been estimated to be 2900 centimorgans (cM; Johnson et al. 1996) and that of the medaka (*Oryzias latipes*) to be 2480 cM (Wada et al. 1995). There currently are efforts to include zebrafish among genome projects of model species sponsored by the National Institutes of Health under the Human Genome Project (Roush 1997). Such a massive effort in zebrafish would provide extremely helpful information for understanding the genome of salmonid fishes.

We expected the pink salmon map in females to be longer than these maps because of the polyploid ancestry of salmonids. However, the linkage map in males will be shorter than in

females because of the reduced recombination rate in male salmonids (Johnson et al. 1987). We anticipate that it will be necessary to map approximately 500 markers to insure that new markers can be assigned to an existing LG with high probability (Van der Beek and Van Arendonk 1993). For example, 99% of all loci in the zebrafish were estimated to be located within 20 cM of a marker on the map based upon an earlier report using 414 markers (Postlethwait et al. 1994).

This project has the following overall specific objectives:

1. Develop several hundred variable DNA markers in pink salmon and test them for Mendelian inheritance.
2. Construct a linkage map based upon joint segregation patterns of the DNA polymorphisms detected in previous objective.
3. Map putative lesions identified in Restoration Study \191A.
4. Test for Mendelian inheritance of markers throughout the genome in progeny of fish exposed to oil. Regions that show aberrant segregation ratios in progeny of fish exposed to oil and normal 1:1 ratios in fish not exposed to oil would be candidates for oil-induced lesions.
5. Test for regions of the genome that are associated with traits of adaptive significance (e.g., marine mortality or run-timing).
6. Test if protein markers (allozymes) are under natural selection such that they may not provide accurate information about the genetic structure and amount of gene flow among populations.

The primary focus in FY 98 will be the completion of Objective 2, the construction of the pink salmon linkage map. We propose to consolidate the number of linkage groups to the number of chromosomes (27; 25 autosomes, the X-chromosome, and the Y-chromosome). We also will begin placing loci used in population genetic analysis of pink salmon (allozymes and microsatellites) onto the map using centromere-linkage analysis of half-tetrads (Johnson et al. 1996). We will also place other loci of special importance onto the map (e.g., growth hormone loci and the major histocompatibility loci (MHC; Katagiri 1996). These loci will be used as landmarks to compare recombination rates in males and females and the linkage maps of odd- and even-year fish (O'Brien et al. 1993)

A secondary focus of FY 98 will be to initiate studies at the Alaska SeaLife Center that will use the linkage map to achieve Objectives 5 and 6.

## B. Methods

### Linkage Map (Objectives 1 & 2)

Our initial map is being built using gynogenetic haploid progeny from individual females. This is the same procedure that has been used to build the zebrafish linkage map (Postlethwait et al. 1994). Stanley (1983) reported that haploid embryos of Atlantic salmon (*Salmo salar*) will develop until just prior to the stage of hatching if development of the eggs is activated by sperm in which the DNA has been inactivated by UV-radiation. We have used this technique routinely with fishes of the genus *Oncorhynchus* (Forbes et al. 1994). This allows us to follow the segregation and linkage relationships in haploid progeny from females. The use of haploid progeny avoids possible difficulties of dominance with some types of DNA markers because recessive alleles are not obscured by their dominant alternatives in haploids (Lie et al. 1994). Our current map is based upon some 181 segregating markers in 94 haploid progeny from a single pink salmon female (number 103) that returned to Armin F. Koernig hatchery in Prince William Sound in August 1995 (Fig. 1).

A useful genetic map should contain genetic markers that are abundant, randomly distributed throughout the genome, highly polymorphic, and readily detectable in many laboratories (Jacob et al. 1995). We began using random amplified polymorphic DNA (RAPDs) markers because they fit these criteria and they have been used successfully in constructing linkage maps in zebrafish and medaka (Johnson et al. 1996; Wada et al. 1995). We have switched to two other types of genetic markers that are superior to RAPDs in this work.

PINES: There are a variety of repetitive DNA elements that are scattered throughout the genome of salmonid fishes. Green and Seeb (submitted) have developed a technique that uses the sequences from SINEs (short interspersed elements) and other DNA elements to detect many DNA polymorphisms. They have called this technique SINE-printing. We have modified this technique using other types of repetitive elements for our mapping study to detect a class of molecular markers that we call PINES (paired interspersed nuclear elements).

Kido et al. (1991) described 3 SINEs in salmonid fishes. They documented the presence of two such elements, HpaI and SmaI, in pink salmon. Spruell and Thorgaard (1996) subsequently reported the presence of the 5' end of the third element, FokI, in pink salmon. Goodier and Davidson (1994) confirmed that salmonids also contain the transposon Tc1, a member of another class of repetitive elements. Both SINEs and transposons occur in high copy number and are believed to be ubiquitously dispersed throughout the genome, making them ideal candidates for genomic mapping efforts.

We have used DNA sequences from SINEs and the transposon Tc1 as PCR primers to generate multiple DNA fragments from a single PCR reaction in pink salmon (Allendorf et al. 1997). The theoretical basis for this procedure is similar to the use of the human SINE AluI to identify human chromosomes in somatic cell hybridization experiments. Primers homologous to one end of the element are oriented such that they initiate DNA synthesis from the end of the element, progressing into the surrounding genomic DNA. A single primer or combinations of primers

may be used to generate multilocus patterns. Greene and Seeb (submitted) used this technique to confirm the parentage of pink salmon fry, demonstrating the potential for including these fragments in our mapping study. We have used five different pairs of PINE primers to detect 67 segregating markers in our reference family (Allendorf et al. 1997).

AFLPs: Amplification fragment length polymorphisms have been used extensively in the construction of genomic maps in plants (Maheswaran et al. 1997; Becker et al. 1995). AFLP analysis consists of three steps (Vos et al. 1995). The first step is the "restriction/ligation" step. Two restriction enzymes are used to cut the genomic DNA into many fragments. Double stranded adapters that are specific to the restriction sites are then ligated onto the fragments. The second step is the "pre-selective amplification". During this step the restriction fragments are amplified using two primers that are specific to the synthetic adapters. Each of these primers includes an additional one base extension into the genomic DNA fragment flanked by the adapters. This step amplifies only DNA fragments with those two bases on either end, reducing the number of DNA fragments available for subsequent amplification. The final step, "selective amplification," uses an aliquot of the pre-selective products as DNA template. Amplification is conducted with primers that are specific to the synthetic adapters with three additional "selective" bases extending into the genomic DNA fragment. The increasing specificity of the primers used to amplify the fragments results in clean, reproducible banding patterns.

The AFLP technique is especially advantageous in this study for two reasons. First, many bands are produced per reaction and, therefore, more scoreable polymorphic loci are produced per unit effort. Second, the selective amplification step uses a subsample of the PCR products of the preamplification. Up to 133 selective amplifications can be completed from a single pre-amplification that originally used only 0.5 µg of genomic DNA. Much more genomic DNA is needed to produce fewer bands using other methods such as RAPDs. This is an important consideration when dealing with the limited amount of tissue available from haploid embryos.

The completion of a full linkage map is a large task. We will try to continue to develop and use as many time and labor saving procedures as possible (Lincoln and Lander 1992; Taylor et al. 1994; Perlin et al. 1994; Archibald 1994). Our initial linkage map is based upon haploid gynogenetic progeny from females, and will be constructed by computer assisted analysis (MapMaker, Lander et al. 1987). We will compare the recombination rates based upon this map to rates of selected pairs of loci in males. The reduced recombination rates in salmonid males means that it will be easier to assign new markers to a LG using male parents. We will test joint segregation of individual markers from different LG's in females to determine if some of these separate LG's in females are linked in males and are therefore syntenic (on the same chromosome).

Differences in meiosis between male and female salmonids have been found in all species that have been examined (Allendorf and Thorgaard 1984; Johnson et al. 1987). There generally is greater recombination in females than in males (Johnson et al. 1987; Allendorf et al. 1994). In addition, only disomic inheritance has been reported in females. However, in males some loci show patterns of segregation that approach those expected with tetrasomic inheritance (Allendorf

and Thorgaard 1984). We will have to test for segregation and linkage in males as well as females because of these sex-specific differences.

### Results to date:

We have assigned 137 of the 181 markers analyzed (76%) to one of 41 linkage groups (Figure 1). These 41 linkage groups have two to seven markers each at an average interval of 9.7 cM (Table 1; Allendorf et al. 1997). The estimated size of the total pink salmon linkage map based on these data is 3,920 cM. This includes 1,328 cM mapped in Figure 1, 504 cM to account for the distance from the end markers to their adjacent telomeres, and 2124 cM in unfilled gaps in the map. The haploid pink salmon genome is approximately 2.72 billion bp (Johnson et al. 1987); thus, we estimate a physical recombination rate of approximately 694 kbp/cM.

Our results are consistent with the maps constructed in zebrafish (Postlethwait et al. 1994; Johnson et al. 1996) and medaka (Wada et al. 1995). The medaka map contains 227 markers, of which 71% have been assigned to a linkage group. This is very similar to the 76% of the markers that have been assigned to a linkage group in our project. Although our current data set of 181 markers is incomplete, our estimate of total map size is close to what we expected on the basis of the polyploid ancestry of salmonids. The zebrafish map is estimated to be 2900 cM (590 kbp/cM) and the medaka is estimated to be 2480 cM (323 kbp/cM).

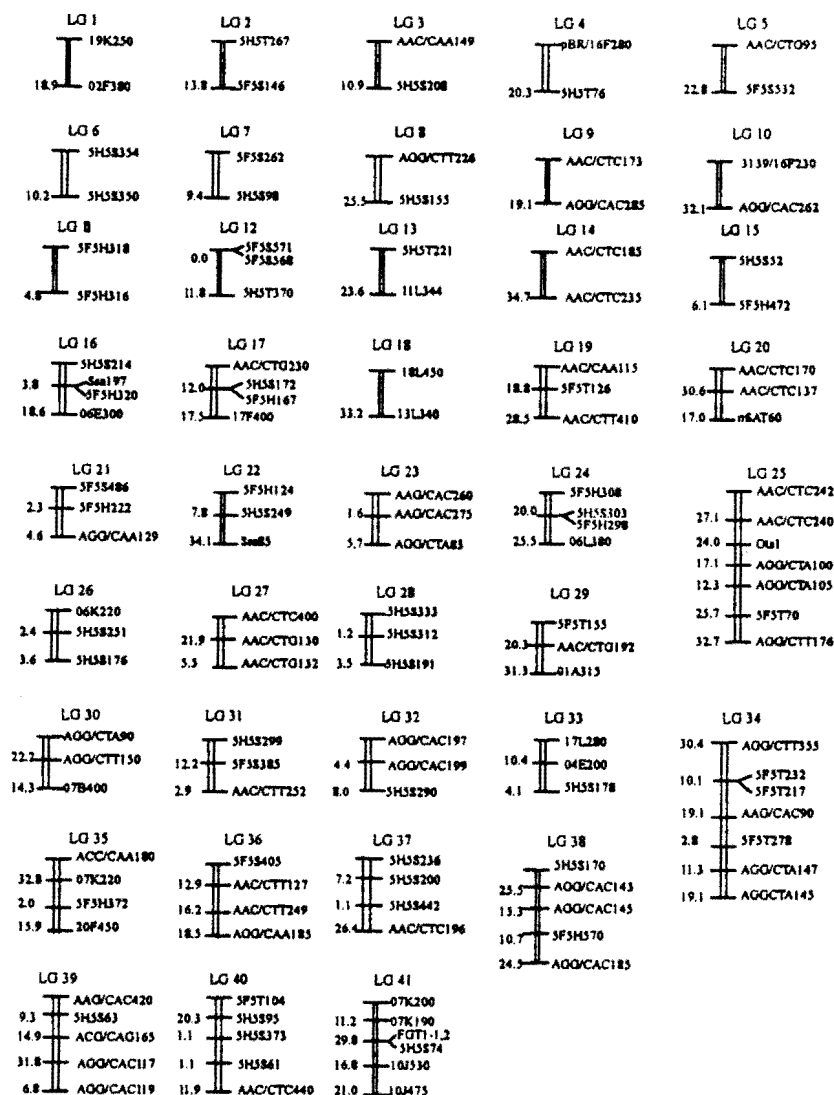
**Table 1. Summary of Pink Salmon Linkage Groups**

Number of markers in linkage group	Number of linkage groups	Average size (cM)	
2	16	18.6	
3	13	24.6	
4	6	38.4	
5	5	57.0	
6	2	85.8	
7	1	138.9	

### Consolidation of the map:

Our primary goal in FY 98 is to consolidate or "close" the pink salmon linkage map. Our current map has 41 linkage groups, while the pink salmon haploid chromosome number in female gametes is 26. Thus, our current map has 15 more linkage groups than chromosomes, and therefore, has at least 15 gaps. We will continue to fill in these gaps by mapping additional loci until we have 26 linkage groups.

Our goal after we consolidate the map will be to place other loci on the map so that the map can be used by other genetic investigators working with pink salmon. The primary types of loci to place on the map will be allozymes and microsatellites that are currently being used in



**Figure 1**

Genetic linkage map of pink salmon. Numbers to the left of the linkage groups indicate recombination rates (centimorgans, cM). To the right of the linkage groups are the locus names.

pink salmon population genetic studies. We will also map other loci that are of special interest and usefulness, e.g., growth hormone loci, and the major histocompatibility complex in pink salmon described by Katagiri et al. (1996).

We will use a procedure using centromere-linkage analysis of half-tetrads developed by Johnson et al. (1996) in zebrafish to consolidate the map and to place these markers on the pink salmon map. This is a very efficient procedure that can place a new locus on the map with a minimum number of analyses. This procedure requires having a marker near the centromere for each of the 26 linkage groups. We have gynogenetic diploid progeny from female 103 that will be used to place the centromeres on our linkage map (Allendorf et al. 1986). The PINE markers may be especially useful in this procedure. Preliminary gene-centromere mapping data suggest these markers tend to map near the centromere (Green and Seeb, manuscript).

We plan to map codominant allozyme and microsatellite loci throughout the genome that can be used as landmarks in mapping studies. These landmark loci will be used to test for differences in the linkage map in odd- and even-year pink salmon. In addition, we will test for differences in recombination rates, crossover interference, and residual tetrasomic inheritance in males and females (Allendorf and Danzmann 1997). We will use the allozyme loci that have already been described in Prince William Sound pink salmon (Restoration Study \196). We will use microsatellite loci that have been described to work in other *Oncorhynchus* species. Over 100 microsatellite loci have been mapped in rainbow trout (Roy Danzmann, personal communication). These loci will be particularly valuable for use in pink salmon and for comparing the maps in pink salmon and rainbow trout.

#### **Identification of the Y-chromosome linkage group:**

We will take a different approach to map the sex-chromosomes in pink salmon. Species in the genus *Oncorhynchus* have an X-Y sex-determining system in which females are XX and males are XY (Allendorf et al. 1994). However, unlike the mammalian X-Y system, most of the Y-chromosome does appear to be functional (i.e., homologous to the X-chromosome). It will be impossible to identify the Y-chromosome using gynogenetic haploids because they do not possess a Y-chromosome. We have previously identified a growth hormone gene that is in the sex-determining region of the Y-chromosome in coho and chinook salmon (Forbes et al. 1994). Additional studies have shown that this gene is actually a non-functional pseudogene (*GH-2p*) and that it is present also in pink and chum salmon (McKay et al. 1996; Du et al. 199; Kavsan et al. 1994; Robert Devlin personal communication). We are using PCR primers to identify the presence of the *GH-2p* pseudogene (Allendorf et al. 1997). This allows us to determine whether or not an individual possesses a Y-chromosome. (PCR primer sequences: left, 5'-tttctctacgtctacattct-3'; right 5'-gtctggctagggtactcca-3' (courtesy R. H. Devlin 1996).

We will screen diploid families of pink salmon for Y-linked markers using bulking or pooling of DNA samples. Pooling DNA to screen for PCR-based markers is both cost-effective and time saving when interested in targeting a particular region of the genome (Giovannoni et al. 1991). DNA will be pooled from males and females that are full-sibs to screen for markers linked to the sex-determining region using AFLP and PINE analysis. We will search for markers that are present in the pooled male samples but not the pooled female samples; this pattern would suggest that the marker is specific to the Y-chromosome. Bands that appear to be sex-specific in bulked analysis will then be examined in individual progeny. We will then map these Y-linked markers

using our linkage map produced with gynogenetic diploids to detect which linkage group is the X-chromosome.

#### Identification and Location of Oil-Induced Lesions (Objectives 3 & 4)

This work will be done in collaboration with efforts to detect oil-induced genetic damage under Component 3 of Restoration Study \191A. Lesions identified in that study through DNA assays of introns, microsatellite loci, or mutational hot spot regions will be tested for joint-segregation with several hundred DNA markers to identify the location of such lesions in the pink salmon genome. A recent paper has found that microsatellite loci show genetic hypermutability because of defects in DNA mismatch repair (Parsons et al. 1995).

Perhaps a more promising approach, however, is to test for regions of the genome associated with non-random survival in haploid progeny. Restoration Study \191A will test for decreased survival in haploid androgens of oil-exposed ancestry. Examining the segregation of markers throughout the genome in these androgens would provide a more powerful test for lesions. Regions of the genome that depart from the expected 1:1 Mendelian ratio would be candidates for lesions. We will also compare Mendelian ratios in haploid gynogens in a similar manner to haploid androgens. The examination of segregation in gynogenetic and androgenetic haploids will also allow testing for oil-induced chromosomal rearrangements (e.g., inversions and deletions).

#### Phenotypic Effects and Fitness (Objectives 5 & 6)

The completion of a genome map for pink salmon will allow us to address important genetic issues related to two other Components of the Pink Salmon Restoration Program. The numerous genetic markers identified in the course of this study will provide greatly increased power and resolution to identify stocks of pink salmon on a very fine scale (Stock Separation and Management). The genetic map will allow us to test for the presence of genes having major effects on phenotypes of importance for the management of pink salmon, and to test for phenotypes associated with specific combinations of multilocus genotypes (Lander and Schork 1994).

This aspect of the research will be performed at the Alaska SeaLife Center Research Facilities. Large numbers of marked fish will be released and then collected when they return to the facility at sexual maturity. A large sample of the fish will be collected at release so that the genetic characteristics of the fish can be described prior to the marine phase of the life cycle. We will test for genetic effects on phenotypes of special importance by comparing the sample of the released fish with the returning fish. This will allow us to test for genes having a major effect on marine survival. We will test for loci or regions of the genome that have a large effect on phenotypes of interest, so-called quantitative trait loci (QTL's).



In addition, previous work has demonstrated genetic differences between early and late run fish, and that differences in run-timing has a genetic basis (Smoker et al. in press). We will compare the genotypes of fish returning to the facility at different times to test for genes having a major effect on run timing. We will use a suite of genetic markers spread uniformly throughout the genome. Regions of the genome that show major associations with run-timing can then be examined in more detail by comparing additional markers within that region. A similar approach using only 10 protein markers in hatchery rainbow trout revealed several regions of the genome associated with time of spawning (Leary et al. 1989)

Karl and Avise (1992) reported concordant patterns of genetic differentiation for mitochondrial DNA and four nuclear DNA loci in the American oyster (*Crassostrea virginica*) along the east coast of North America. In contrast, previous allozyme studies had not detected these genetic differences among these same populations. Karl and Avise concluded that the pattern observed for the DNA markers reflected the historical patterns of isolation and gene flow among these populations while this pattern is obscured in the allozymes because of "balancing selection" at the allozyme loci. Similar results have been reported recently in the Atlantic cod (Pogson et al. 1995). These results provide an important challenge to the generally accepted utility of allozyme markers for describing historical patterns and amounts of gene flow between populations. That is, if allozymes are under strong natural selection then they may not provide accurate information about the genetic structure and amount of gene flow among populations.

Pink salmon that are more heterozygous at allozyme loci have greater viability and growth rates than more homozygous individuals (Altukhov et al. 1991; Zhivotovsky et al. 1987). Similar results have been reported in other salmonid species for many phenotypes of evolutionary importance (e.g., developmental rate, egg size, and disease resistance; reviewed by Ferguson 1992). Positive associations between heterozygosity at allozyme loci and important phenotypic characters, such as growth rate, survival, fertility, disease resistance, developmental rate, and developmental stability, have been described in many organisms (reviewed by Zouros and Foltz 1986; Allendorf and Leary 1986).

The mechanism underlying these associations remains unknown. The possible explanations most often considered are either the associations are the consequence of heterozygosity at the loci examined, or the loci examined may be in linkage disequilibrium with other loci that affect the traits being studied (Leary et al. 1987). It has been argued that these relationships between multiple locus heterozygosity and phenotypes have been found with allozymes because these loci are important in ATP production and protein catabolism (Koehn et al. 1988). We propose to distinguish between these hypotheses by comparing the effects on marine survival of DNA markers and protein polymorphisms. If the enzyme loci themselves are responsible for this effect, then we would expect to find an association between enzyme genotypes and survival, but not between genotypes at DNA markers spread throughout the nuclear genome.

We propose to release full-sib families from 50-100 females that have each been mated with a different male. Each female has approximately 1,500 eggs and we expect survival to return to be approximately 2-9% (J. Seeb, personal communication). This would result in approximately 30-135 returning fish per full-sib family. Experimental fish will be marked with an adipose fin clip;

returning fish will be assigned to individual families on the basis of their genotype at the landmark loci spread throughout the nuclear genome and mtDNA.

This is an extremely powerful experimental design that will allow us to measure a multitude of parameters for the first time with pink salmon or any salmonid fish. The most powerful aspect of this experiment will be the capability of measuring fitness for loci spread throughout the genome. In the case of males, fitness will be estimated by survivorship (viability) from egg to return at sexual maturity. In the case of females, we will use both survivorship and the number of eggs produced so that we can take into account both viability and fecundity.

### **C. Cooperating Agencies, Contracts, and Other Agency Assistance**

None anticipated at this time.

## **SCHEDULE**

### **A. Measurable Project Tasks for FY 98 (1 Oct 97 - 30 Sep 98)**

1 Oct 97 - 30 Sep 98:	Continued screening of DNA polymorphisms to test for Mendelian inheritance and joint segregation in 1995 brood-year progeny. We plan to have consolidated the map by the end of this time period.
1 Apr 98 - Sep 98	Place allozyme, microsatellite, and other codominant markers (MHC, etc.) on to the map.
1 Jul 98 - 30 Sep 98:	Begin studies at the Alaska SeaLife Center to test for adaptive significance and major phenotypic effects of the loci in the pink salmon genome.

### **B. Project Milestones and Endpoints**

Objective 1: This objective has been completed.

Objective 2: This objective will be completed by the end of year 3 (FY 98).

Objective 3: This objective will be completed by the end of year 5.

Objective 4: This objective will be completed by the end of year 5.

Objective 5: This objective will be completed by the end of year 5.

Objective 6: This objective will be completed by the end of year 5.

### **C. Completion Date**

We propose to continue this work for five years. This will allow us to complete multigenerational studies of inheritance with pink salmon. New genetic markers have been developed in the first year of the study. However, it will take several years to map the markers in both males and females in both odd- and even-year fish. Different objectives will be met throughout the course of the research. This project would be carried out in collaboration with Dr. James E. Seeb, Alaska Department of Fish and Game. The primary laboratory aspects of this research would be carried out at the University of Montana. We also propose to use the Alaska SeaLife Center Research Facilities. This facility will greatly strengthen genetic investigations with pink salmon by allowing multigenerational studies. We have not included budget costs associated with the Alaska SeaLife Center facility because we have been informed by Dr. Michael Castellini that such fees should be left out of the budget and they will be negotiated later.

### **PUBLICATIONS AND REPORTS**

Allendorf, F. W., P. Spruell, K. L. Knudsen, K. R. Lindner and K. L. Pilgrim 1997.  
Construction of a Linkage Map for the Pink Salmon Genome, Exxon Valdez Oil Spill  
Restoration Project Annual Report (Restoration Project 97190), University of Montana,  
Missoula, Montana.

The following manuscript is nearing completion and will be submitted soon:

Spruell, P., B. A. Greene, C. Habicht, K.L. Knudsen, K. R. Lindner, K. L. Pilgrim, J. E.  
Seeb, and F. W. Allendorf. (In preparation). Inheritance of nuclear DNA markers in  
haploid pink salmon embryos. *Molecular Ecology*.

We anticipate writing and submitting a manuscript by the end of year three that will describe the pink salmon linkage map to the journal *Genetics*.

### **PROFESSIONAL CONFERENCES**

We anticipate presenting our results at professional and scientific meetings based on our results. We do not know at present the specifics of these presentations in FY 98.

### **COORDINATION AND INTEGRATION OF RESTORATION EFFORT**

This work is being done in collaboration with James E. Seeb, Principal Geneticist, ADFG. The inheritance experiments will be in coordination with the project Oil-Related Embryo Mortalities (Restoration Study \191A). Dr. Seeb and I are also coordinating plans to use the Alaska SeaLife

Center Research Facilities at Seward. Where possible we will share fish samples, gametes, laboratory equipment, and fish rearing facilities.

This work is related to my ongoing genetic research with salmonid fishes that has been supported by the National Science Foundation since 1980. Many of the techniques and approaches proposed here are based upon the results of that research. I also intend to continue seeking support from NSF that will complement the research proposed here. A genetic map for pink salmon will allow us to address a number of fundamental questions in the conservation and genetics of pink salmon and other *Oncorhynchus* species.

## **EXPLANATION OF CHANGES IN CONTINUING PROJECTS**

The changes in this proposal reflect the more rapid than anticipated progress and the review of our FY 97 proposal that said we must seek cost sharing from other sources.

## **PROPOSED PRINCIPAL INVESTIGATOR**

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## PRINCIPLE INVESTIGATOR

FRED W. ALLENDORF: Principal Investigator

BIRTH: 29 April 1947; Philadelphia, Pennsylvania

MILITARY SERVICE: U.S. Army, 1965-1968 (Vietnam, 1966-1967)

EDUCATION: B.S., Zoology, Pennsylvania State University, 1971  
M.S., Fisheries, University of Washington, 1973  
Ph.D., Genetics and Fisheries, University of Washington, 1975 (co-directors,  
Joe Felsenstein and Fred Utter)

## POSITIONS:

1975-1976 Lektor, Department of Genetics and Ecology, Aarhus University, Denmark  
1976-1979 Assistant Professor of Zoology, University of Montana  
1978-1979 NATO Fellow, Genetics Research Unit, University of Nottingham, England  
1979-1984 Associate Professor of Zoology, University of Montana  
1983-1984 Visiting Scientist, Department of Genetics, Univ. of California, Davis  
1984-1989 Professor of Zoology, University of Montana  
1989-1990 Program Director, Population Biology and Physiological Ecology, National  
Science Foundation (NSF)  
1992-1993 Visiting Professor, University of Oregon  
1990- Professor of Biology, University of Montana  
1993-1996 Director, Organismal Biology and Ecology Graduate Program, University of  
Montana

HONORS: NATO/NSF Postdoctoral Fellowship, University of Nottingham, 1978-1979  
European Molecular Biology Organisation (EMBO), Fellowship, University of  
Stockholm, 1979  
Distinguished Scholar Award, University of Montana, June 1985  
Burlington Northern Faculty Achievement Award for Research, University of Montana,  
June 1987  
Elected Fellow, American Association for the Advancement of Science (AAAS), February  
1987  
Burlington Northern Faculty Achievement Award for Research, University of Montana,  
May 1991  
Elected Member, AAAS Council (Biological Sciences Division)

## MAJOR GRANTS:

National Science Foundation Research Grant, EPSCR, 1980-1983, \$70,000  
National Science Foundation Research Grant, Population Biology, 1980-1982, \$60,000  
National Science Foundation Research Grant, 1983-1986, \$121,000

National Science Foundation, Faculty Research Opportunity Award, 1986, \$10,000  
United States Department of Agriculture Grant, Aquaculture, 1983-1985, \$43,000  
National Science Foundation Research Grant, 1986-1989, \$148,000  
National Science Foundation, Dissertation Research Grant, 1988-1990, \$9,850  
National Science Foundation Research Grant, 1989-1993, \$150,000  
National Science Foundation Research Grant, Conservation and Restoration Biology,  
1993-1996, \$250,000

ASSOCIATE EDITORSHIPS: Evolution (1987-1990)  
Journal of Heredity (1986-1989)  
Progressive Fish Culturist (1986-1989)  
Molecular Biology and Evolution (1994-1996)

EDITORIAL BOARDS: Molecular Biology and Evolution (1983-1989)  
Conservation Biology (1990-1993)  
Molecular Ecology (1991-present)

PROFESSIONAL SERVICE:

Panel Member, Population Biology and Physiological Ecology, NSF (1987-1989)  
Panel Member, International Program, National Science Foundation (1987)  
Panel Member, Conservation and Restoration Biology, NSF (1991-1992; 1995)  
Council Member, The American Genetic Association (1986-1989)  
Genetics Nomenclature Committee, American Fisheries Society (1986-present)  
Member, Committee on the Protection and Management of Pacific Northwest Anadromous  
Salmonids, National Research Council (1992-present)  
Chair, Committee of Visitors, Systematic and Population Biology Programs, NSF (1993)

PROFESSIONAL SOCIETIES: Society for the Study of Evolution  
American Society of Naturalists  
Genetics Society of America  
Society for Conservation Biology  
American Association for the Advancement  
of Science  
American Society of Ichthyologists and Herpetologists  
American Fisheries Society  
American Genetic Association  
Desert Fishes Council  
Ecological Society of America  
Montana Native Plant Society  
Society of Systematic Biologists  
Society for Molecular Biology and Evolution

## BOOK CHAPTERS:

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## OTHER KEY PERSONNEL

Paul Spruell will be responsible for direct oversight of all the activities in the laboratory and for statistical analysis. Kate Lindner will be responsible for all laboratory procedures (extraction of DNA, PCR analysis, and gel electrophoresis).

*Paul Spruell*: Research Scientist

BORN: August 14, 1965 Bloomington, IL USA

EDUCATION:

B.S.. Ecology, Ethology and Evolution, University of Illinois 1987



M.S. Fisheries and Wildlife, Michigan State University 1989  
Ph.D. Zoology, Washington State University 1994

## AWARDS

Guy Brislawn Award for the Outstanding WSU Zoology graduate student. 1994.

## SOCIETIES

American Fisheries Society 1987-present  
American Society of Ichthyologists and Herpetologists 1993-present

## RESEARCH INTERESTS

Conservation genetics of fishes  
Population genetics of fishes  
Alternate reproductive strategies in fishes  
Application of molecular tools to conservation and management  
Evolutionary biology and systematics of fishes

## POSITIONS

Post-doctoral Research Associate, University of Montana	08/95-present
Post-doctoral Research Associate, Washington State University	11/94-08/95
Post-doctoral Research Associate, S.U.N.Y-Stony Brook	08/94-11/94
Graduate Assistant, Washington State University	08/89-08/94
Graduate Assistant, Michigan State University	06/83-08/89

## PUBLICATIONS:

### Dissertations and Thesis:

Spruell, P. 1989. Evaluation of triploid induction in chinook salmon using microwave radiation and growth comparisons of diploid and triploid chinook salmon. M.S. thesis. Michigan State University.

Spruell, P. 1994. DNA fingerprinting of fishes using tandemly repeated and interspersed DNA sequences. Ph.D. dissertation. Washington State University.

### Primary Literature:

- Spruell, P., S. A. Cummings, Y. Kim, and G. H. Thorgaard. 1994. Comparison of three anadromous rainbow trout populations using DNA fingerprinting and mixed DNA samples. *Can. J. Fish. Aquat. Sci.* 51 (Suppl. 1): 252-257.
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# 1998 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1997 - September 30, 1998

Budget Category:	Authorized FY 1997	Proposed FY 1998							
Personnel		\$0.0							
Travel		\$0.0							
Contractual		\$197.8							
Commodities		\$0.0							
Equipment		\$0.0							
Subtotal	\$0.0	\$197.8	LONG RANGE FUNDING REQUIREMENTS						
General Administration		\$13.8		Estimated FY 1999	Estimated FY 2000	Estimated FY 2001	Estimated FY 2002		
Project Total	\$0.0	\$211.6		\$187.0	\$187.0				
Full-time Equivalents (FTE)		0.0							
Dollar amounts are shown in thousands of dollars.									
Other Resources									
Comments:									

**1998**

Project Number: 98190  
 Project Title: Construction of a Linkage Map for the Pink Salmon  
 Genome  
 Agency: Alaska Department of Fish & Game

FORM 3A  
 TRUSTEE  
 AGENCY  
 SUMMARY

# 1998 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1997 - September 30, 1998

Personnel Costs:		GS/Range/ Step	Months Budgeted	Monthly Costs	Overtime	Proposed FY 1998
Name	Position Description					
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
Subtotal			0.0	0.0	0.0	
<b>Personnel Total</b>						<b>\$0.0</b>

Travel Costs:		Ticket Price	Round Trips	Total Days	Daily Per Diem	Proposed FY 1998
Description						
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
<b>Travel Total</b>						<b>\$0.0</b>

**1998**

Project Number: 98190  
 Project Title: Construction of a Linkage Map for the Pink Salmon  
 Genome  
 Agency: Alaska Department of Fish & Game

FORM 3B  
 Personnel  
 & Travel  
 DETAIL



**1998 EXXON VALDEZ TRUSTEE NCIL PROJECT BUDGET**  
October 1, 1997 - September 30, 1998

<b>Contractual Costs:</b>		Proposed
Description		FY 1998
Contract with the University of Montana		197.8
When a non-trustee organization is used, the form 4A is required.		
<b>Contractual Total</b>		\$197.8
<b>Commodities Costs:</b>		Proposed
Description		FY 1998
<b>Commodities Total</b>		\$0.0

## 1998

Project Number: 98190  
Project Title: Construction of a Linkage Map for the Pink Salmon Genome  
Agency: Alaska Department of Fish & Game

FORM 3B  
Contractual &  
Commodities  
DETAIL

October 1, 1997 - September 30, 1998

1998

### FORM 3B Equipment DETAIL

# 1998 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1997 - September 30, 1998

Budget Category:	Authorized FY 1997	Proposed FY 1998						
Personnel		\$108.5						
Travel		\$11.3						
Contractual		\$0.0						
Commodities		\$42.7						
Equipment		\$0.0	LONG RANGE FUNDING REQUIREMENTS					
Subtotal	\$0.0	\$162.5		Estimated FY 1999	Estimated FY 2000	Estimated FY 2001	Estimated FY 2002	
Indirect		\$35.3						
Project Total	\$0.0	\$197.8		\$175.0	\$175.0	\$0.0	\$0.0	
Full-time Equivalents (FTE)		54.0						
Dollar amounts are shown in thousands of dollars.								
Other Resources								
<p>Comments:</p> <p>Indirect cost is based on The University of Montana rate of 43.7% of salaries and wages.</p> <p>The cost of preparing a report of the FY 98 activity is included.</p>								

**1998**

Project Number: 98190  
 Project Title: Construction of a Linkage Map for the Pink Salmon  
 Genome  
 Name: University of Montana

FORM 4A  
 Non-Trustee  
 SUMMARY

# 1998 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1997 - September 30, 1998

Personnel Costs:			Months	Monthly	Overtime	Proposed
Name	Position Description		Budgeted	Costs		FY 1998
F. Allendorf	Project Director		3.0	7.16	0.0	21.5
P. Spruell	Research Scientist		12.0	2.79	0.0	33.5
K. Lindner	Research Assistant		12.0	2.14	0.0	25.7
						0.0
						0.0
						0.0
	Fringe Benefits		27.0	1.03		27.8
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
Subtotal			54.0	13.1	0.0	
Personnel Total						\$108.5
Travel Costs:			Ticket	Round	Total	Proposed
Description			Price	Trips	Days	FY 1998
Missoula to Anchorage for Trustee Council Annual Restoration Workshop, research collaboration with ADFG, and initiation of studies at the ASLC			0.65	8	40	9.2
						0.0
						0.0
Professional and Scientific meetings			0.75	2	6	2.1
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
Travel Total						\$11.3

1998

Project Number: 98190  
 Project Title: Construction of a Linkage Map for the Pink Salmon Genome  
 Name: University of Montana

FORM 4B  
 Personnel  
 & Travel  
 DETAIL

Prepared: 6 of 8

4/15/97

## 1998 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1997 - September 30, 1998

<b>Contractual Costs:</b>		Proposed
Description		FY 1998
<b>Contractual Total</b>		<b>\$0.0</b>
<b>Commodities Costs:</b>		Proposed
Description		FY 1998
Materials and supplies for PCR analysis		26.5
Communications		0.7
FMBIO fluorescent scanner service contract		8.0
Equipment repair and maintenance		7.5
<b>Commodities Total</b>		<b>\$42.7</b>

## 1998

Project Number: 98190  
Project Title: Construction of a Linkage Map for the Pink Salmon Genome  
Name: University of Montana

FORM 4B  
Contractual &  
Commodities  
DETAIL

Prepared: 7 of 8

4/15/97

# 1998 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1997 - September 30, 1998

New Equipment Purchases:		Number of Units	Unit Price	Proposed FY 1998
Description				
				0.0
				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 a <b>New Equipment Total</b>				\$0.0
Existing Equipment Usage:		Number of Units		
Description				
Hitachi FMBIO Fluorescent Imaging Scanner		1		

**1998**

Project Number: 98190  
 Project Title: Construction of a Linkage Map for the Pink Salmon  
 Genome  
 Name: University of Montana

FORM 4B  
 Equipment  
 DETAIL



## Field Examination of Oil-Related Embryo Mortalities in Pink Salmon Populations in Prince William Sound

**Project Number:** 98191A

**Restoration Category:** Research and Monitoring

**Proposer:** Alaska Department of Fish and Game

**Lead Trustee Agency:** Alaska Department of Fish and Game

**Cooperating Agencies:** None

**Alaska SeaLife Center:**

**Duration:** 9th year, 10- year project

**Cost FY 98:** \$164,200

**Cost FY 99:** \$ 58,700

**Cost FY 00:** \$ 00.0

**Cost FY 01:** \$ 00.0

**Cost FY 02:** \$ 00.0

**Geographic Area:** Prince William Sound

**Injured Resource/Service:** Pink Salmon

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TRUSTEE COUNCIL

### ABSTRACT

Elevated embryo mortalities were detected in populations of pink salmon inhabiting oiled streams. These increased rates of mortality persisted annually through the 1993 field season, three generations after the oil spill, suggesting that damage may have occurred as a result of exposure to oil during early developmental life-stages. The consequences of this putative damage include physiological dysfunction of individuals and reduced reproductive capacity of wild pink salmon populations. The 1994, 1995, and 1996 field results show no statistical difference in embryo mortality between oil-contaminated and reference streams. The purpose of this project is to continue to monitor the recovery of pink salmon embryos in the field. If there is again no difference in embryo mortality between oil-contaminated and reference streams, this project will be closed out in FY99.



## INTRODUCTION

Elevated embryo mortalities were detected in populations of pink salmon *Oncorhynchus gorbuscha* inhabiting oiled streams. These increased rates of mortality persisted annually through the 1993 field season, three generations after the oil spill, suggesting that damage may have occurred as a result of exposure to oil during early developmental life-stages. The consequences of this putative damage include physiological dysfunction of individuals and reduced reproductive capacity of wild pink salmon populations.

The purpose of this study is to continue to monitor the recovery of pink salmon embryos in the field. In this study we will survey the same streams examined during the Natural Resource Damage Assessment (NRDA) studies.

## NEED FOR THE PROJECT

### A. Statement of the Problem

Pink salmon embryos that incubated in the oiled intertidal spawning areas in Prince William Sound (PWS) in 1989, 1990, 1991, 1992, and 1993 appear to have been adversely affected. Oil was deposited in layers of varying thickness in the intertidal portions of streams utilized by spawning pink salmon. Pink salmon eggs deposited in 1988 (1988 brood year) emerged as fry through the oiled spawning gravels during the spring of 1989 and began feeding on oiled plankton. These fish showed decreased growth due to oiling (Willette et al. 1994). Although gross oil levels decreased during the summer of 1989, contamination in the intertidal zone was still evident. The pink salmon eggs deposited during the late summer of 1989 (the 1989 brood year) were exposed to intra-gravel contamination from late August 1989 through mid-May 1990. Sharr et al. (1994a) and Bue et al. (1996) detected elevated mortalities of pink salmon embryos in the intertidal zones of oiled streams while no difference between oiled and non-oiled streams was detected above mean high tide. Elevated embryo mortalities in oiled streams were again detected in the 1990 brood year, but only in the highest intertidal spawning zone (Sharr et al. 1994a; Bue et al. 1996). Visual observations indicated that the majority of the remaining oil was deposited in this zone. Spawning areas lower in the intertidal zone seemed to be recovering as embryo mortalities in these areas were not statistically different from non-oil impacted streams.

Surprisingly, Sharr et al. (1994a) and Bue et al. (1996) found increased embryo mortalities in oiled streams during the 1991 fall survey. Furthermore, significant differences in embryo mortality occurred at all tidal zones, including the area above mean high tide. Clearly, the elevated embryo mortalities in the oiled streams were not the direct effect of recent oiling. The 1991 adult returns were the progeny of the 1989 brood year, the group with the highest exposure to intra-gravel oil (the 1989-90 incubation period). We hypothesize that the elevated embryo mortalities in 1991 may be the result of damage acquired during embryonic development. Elevated embryo mortalities at all tidal zones in oiled streams were again detected during the 1992 survey (Sharr et al. 1994b; Bue et al. 1996). A hatchery incubation experiment using gametes from fish returning to oiled and control streams in 1993 indicate that mortality differences observed during past studies cannot be attributed to environmental factors or sampling design (Sharr et al. 1994c).

The aggregate of evidence from the field studies and incubation experiment suggests that the embryos exposed to oil in 1989 and 1990 accumulated deleterious mutations in the germline. This hypothesis of genetic damage is consistent with previous field observations and laboratory experiments on the effects of crude oil on early life stages of fish. Long term intra-gravel oil exposures (7-8 months) to freshly fertilized eggs provide embryos sufficient time to accumulate polynuclear aromatic hydrocarbons (PAH's) from very low aqueous concentrations of crude oil. PAH's are abundant in crude oil and are potent clastogens (i.e. capable of breaking chromosomes). Mironov (1969) observed reduced survival of fish embryos and larvae exposed to very low aqueous doses (1 ul oil/l seawater) of oil. Longwell (1977) reported genetic damage in pelagic embryos affected by the *Argo Merchant* oil spill. Moles et al. (1987) confirmed that pink salmon embryos take up PAH's and demonstrated that the uptake was much greater in an intertidal environment than in strictly freshwater conditions. Biggs et al. (1991) found greater numbers of chromosome aberrations in larval herring which incubated in oiled areas than in non-oiled areas. It is logical that the same type of damage may have occurred in pink salmon, and this damage could have affected the germline of exposed individuals (cf., Malkin 1994).

Genetic damage induced by genotoxins can be classified into two general categories: small changes to nucleotide sequence caused by base substitutions, deletions, or additions (microlesions); and changes in chromosome structure through inversions, larger scale deletions, or translocations (macrolesions). Increasing concern about the effects of chemicals in the environment has lead to a proliferation of assays developed to assess their genotoxic potential (reviewed in Landolt and Kocan 1983, Kocan and Powell 1985, Liguori and Landolt 1985). Because chemical agents that induce mutations in DNA are also likely to produce cytologically recognizable chromosome damage expressed as structural changes or "aberrations" (Evans 1976), cytogenetic techniques can be used to detect these kinds of damage. Alternatively, microlesions may be detected by exposing detrimental recessive alleles through haploid androgenesis (Armstrong and Fletcher 1983) or by directly examining the base-pair structure of the DNA molecule (e.g., Orita et al. 1989a, 1989b; Hovig et al. 1991).

## **B. Rationale/Link to Restoration**

In this project we propose to continue monitoring embryo mortality rates in oiled and reference streams. This study will provide resource managers with information about the magnitude and persistence of damages sustained by wild pink salmon due to the *Exxon Valdez* oil spill (EVOS). Efforts to restore damaged pink salmon populations depend upon the ability of fishery managers to identify sources of reduced survival and to monitor their persistence. The potential of long term oil exposures to cause damage needs to be understood so that spawning escapement goals can be adjusted if necessary to effect recovery.

## **C. Location**

Embryo sampling in PWS will be conducted in the fall on 31 streams (Figure 1). These same 31 streams have been sampled annually since 1989.

## **COMMUNITY INVOLVEMENT**

Laboratory analyses and reporting are technical pursuits that will be conducted by or supervised by professional scientists. Wherever possible, local-hire will be used to fill field positions required for sampling or for routine laboratory positions. People from the communities in PWS will have an opportunity to participate in this project as employees of the ADF&G which gives local residents priority in hiring for state employment.

## **PROJECT DESIGN**

### **A. Objectives**

The purpose of this project is to monitor the recovery of damaged pink salmon populations in oil-contaminated streams. Working objectives are:

1. Estimate the density, by tidal zone, of embryos in 31 streams using counts of live and dead embryos.
2. Estimate mortality of pink salmon embryos in both oil-contaminated streams and noncontaminated reference streams.

### **B. Methods**

#### **a. Data Collection**

Embryo sampling will be conducted from late September to mid-October in 31 streams (Figure 1). Embryo development by this time includes stages from uneyed embryo through recently hatched fry. The streams were selected using the following criteria:

- (1) Adult salmon returns were adequate to support a high probability of success in embryo sampling.
- (2) Embryo sampling had been done in past years.
- (3) Streams with low to no oil impact, i.e., reference streams, were selected in the immediate vicinity of high oil impact streams to control for possible variability in embryo survival due to environmental conditions.

Twenty eight of the 31 streams are located in the western half of PWS in close geographic proximity to each other and in the area where oil impacts were greatest. Twelve experienced impacts ranging from light to heavy oiling. Most of the streams which sustained suspected or obvious oil impact were not sampled for embryos or fry prior to the EVOS. Among the 12 streams where oil was visibly present in 1989, only one had a history of embryo sampling.

Methods for embryo sampling were modeled after procedures described by Pirtle and McCurdy (1977). On each study stream, four zones, three intertidal and one above most tidal influence, were measured from the mean low tide mark using computer generated tide tables and a surveyors level. Boundaries between zones were marked with stakes. The four zones were: 1.8-2.4 m, 2.4-3.0 m, 3.0-3.7 m above mean low water, and upstream of mean high tide (3.7 m). A linear transect 30.5 m in length was established for embryo samples in each zone. The transect ran diagonally across the stream. To insure continuity of transects between years, transect locations were marked with stakes and carefully photographed from at least two perspectives. Fourteen 0.186 m<sup>2</sup>, circular digs were systematically made along each transect using a high pressure hose to flush embryos from the gravel. Embryos and fry were caught in a specially designed net.

The following data will be collected for each tide zone transect during embryo sampling:

- (1) The sample date.
- (2) The sample tide zone.
- (3) The start and stop time for each tide zone transect.
- (4) Numbers and condition (live or dead) of embryos by species.
- (5) A subjective estimate of the overall percent yolk sac absorption for fry.

Data will be transferred from field notebooks into a Lotus spreadsheet for editing and summarizing.

Pink salmon embryos will be separated from chum *O. keta* and coho *O. kisutch* salmon embryos by their smaller size. Chum salmon embryos will be separated from coho salmon embryos by their greater development and different coloration. An embryo will be considered dead if it is opaque or discolored with coagulated lipids. Sampling often kills fry (especially newly hatched fry), so fry will only be considered dead if decomposition is evident.

#### b. Data Analysis

Numbers of live and dead embryos and fry will be summarized by date, stream, level of hydrocarbon impact, and stream zone. Densities of live embryos for stream *i*, zone *j* in m<sup>2</sup> ( $\hat{E}_{ij}$ ) will be estimated by:

$$\hat{E}_{ij} = \frac{\sum LE_{ijk}}{0.3n_{ij}}, \quad (1)$$

where  $LE_{ijk}$  is the number of live embryos found in the  $k^{\text{th}}$  dig, in stream *i*, zone *j*, and  $n_{ij}$  is the number of digs from stream *i*, zone *j*. Densities of dead embryos will be calculated using the same estimator with appropriate substitutions.

Pink salmon embryo mortality will be estimated using the following relationship:

$$\hat{M}_{ij} = \frac{\Sigma(DE_{eijk} \quad DF_{eijk})}{\Sigma(LE_{eijk} \quad DE_{eijk} \quad LF_{eijk} \quad DF_{eijk})}, \quad (2)$$

where  $DE_{eijk}$ ,  $DF_{eijk}$ ,  $LE_{eijk}$ , and  $LF_{eijk}$  are the number of dead embryos, dead fry, live embryos, and live fry for the  $k^{\text{th}}$  dig from stream  $i$ , zone  $j$ , collected during embryo dig  $e$ , respectively.

Differences in embryo mortality will be examined using a mixed effects two-factor experiment with repeated measures on one factor (Neter et al. 1990):

$$Y_{ijk} = \mu + O_i + Z_j + (OZ)_{ij} + S_{k(i)} + e_{(ijk)}. \quad (3)$$

The two treatments will be level of oiling, ( $O_i$ , 2 levels; oiled and non-oiled), and height in the intertidal zone ( $Z_j$ , 4 levels; 2.1, 2.7, and 3.4 m above mean low water, and upstream) both fixed effects. The data will be blocked by stream ( $S_{k(i)}$ ), a random effect nested within level of oiling. The interaction of level of oiling and height in the intertidal zone will also be examined. Equality of variances will be tested using the  $F_{\text{max}}$ -test (Sokal and Rohlf, 1981), while normality will be visually assessed using normal quantile-quantile and box plots (Chambers et al. 1983). If the distribution of residuals appears to be non-normal, data transformations will be examined. If a significant difference due to oiling is detected ( $\alpha = 0.05$ ), four contrasts (oil vs. non-oiled for the four stream zones) and corresponding Bonferroni family confidence intervals ( $\alpha = 0.10$  overall) will be estimated.

Extent of oiling for analysis will be based on visual observations of streams (NRDA F/S Study 1 and 2) and hydrocarbon results from mussel samples (NRDA F/S Study 1).

### C. Cooperating Agencies, Contracts, and Other Agency Assistance

The Alaska Department of Fish and Game will be completing all work on this project.

## SCHEDULE

### A. Measurable Project Tasks for FY 98 (October 1, 1997 - September 30, 1998)

Monitoring of Injury to Pink Salmon Embryos in Prince William Sound

15 Sep - 30 Oct 1997: Embryo deposition sampling.

30 Oct 1997 - 30 Mar 1998: Analysis of brood year 1997 embryo data and completion of firstdraft of 97191A report.

15 April 1998 Submit 9719A annual report.

### B. Project Milestones and Endpoints

Annual review: terminate project component if embryo mortalities are not significantly different between oiled and non-oiled study sites for two consecutive years for both the odd- and two-even broodlines

### **C. Completion Date**

The population monitoring components of this study should be continued until the methodology used to monitor is unable to detect a difference in pink salmon embryo mortality between oil contaminated and reference streams. Results to date indicate that recovery is likely ongoing. However, we recommend that this project continue until both odd- and even-broodline pink salmon exhibit no difference in embryo mortality between oiled and non-oiled study sites for two consecutive years based upon the statistical tests described.

### **PUBLICATIONS AND REPORTS**

Field activities will continue for two generations past when injury to salmon embryos and fry can no longer be detected. Until field activities cease, the main product from this project will be an annual report which summarizes the results of the current-year embryo data. The most significant information on damages demonstrated in 1989 through 1992 were presented in a close-out reports for NRDA Study #2 and Restoration Studies R60C and 93003. These results will also be published in a peer-reviewed journal. When restoration field work is complete, a follow up journal article may be appropriate if there have been findings which add significantly to or alter results reported from the NRDA study. An annual project report for FY 98 will be submitted by April 15, 1999.

## **PROFESSIONAL CONFERENCES**

Travel funds have been requested for this project to attend meetings with personnel in Anchorage and Juneau. Two professional conferences are also planned for 1998, the Pink and Chum Workshop, and the state American Fisheries Society meeting. The place and time for these meeting has not been set at this time.

## **NORMAL AGENCY MANAGEMENT**

The Alaska Department of Fish and Game did not fund this research prior to the 1989 *Exxon Valdez* Oil Spill and has no plans to continue funding after recovery is complete. In the past an embryo mortality study was implemented with federal disaster funds following the 1964 earthquake and continued until 1975 when recovery was complete. Continued monitoring of this resource is necessary to document complete recovery.

## **COORDINATION AND INTEGRATION OF RESTORATION EFFORT**

The field data collection for Component 1 of this project is very specific to individual wild pink salmon streams and occurs after most field activities of SEA (97320) and other pink salmon related projects each year. Consequently extensive coordination of field activities is not feasible. However, the vessel used by this project does collect physical and biological oceanographic data for the ADFG, PWSAC, and University of Alaska, and these data will be utilized by several SEA studies.

Final edited data from both components of this project will be stored electronically as computer databases, and final versions will be provided annually to the Information Modelling portion of SEA for incorporation into a centralized ecosystem database.

## **EXPLANATION OF CHANGES IN CONTINUING PROJECTS**

Due to the recovering status of pink salmon, the laboratory verification of the field results was removed from this project in FY96.

## PERSONNEL

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### A. Mark Willette, Area Research Biologist

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Commercial Fisheries Management and Development Division  
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PROJECT RESPONSIBILITIES: Design and supervision of embryo surveys, analysis, report writing

EDUCATION: 1985 Master of Science, Fisheries Oceanography, University of Alaska Fairbanks  
1983 Bachelor of Science, Fisheries Science, University of Alaska Fairbanks

### PROFESSIONAL EXPERIENCE:

1995 - present	Area Research Biologist, Cordova, Commercial Fisheries Management and Development, ADFG
1991 - 1995	Area Resource Development Biologist, Cordova, Commercial Fisheries Management and Development, ADFG
1986 - 1991	Fisheries Instructor/ Assistant Research Professor, School of Fisheries & Ocean Sciences, University of Alaska Fairbanks
1983 - 1985	Research Assistant, School of Fisheries & Ocean Sciences, University of Alaska Fairbanks
1978 - 1983	Fish and Wildlife Technician, Commercial Fisheries Division, ADFG

### SELECTED PUBLICATIONS:

Willette, T.M. 1995 Impacts of the Exxon Valdez Oil Spill on the migration, growth, and survival of juvenile pink salmon in Prince William Sound. In: Proceedings of the Exxon Valdez Oil Spill Symposium, American Fisheries Society Symposium Series, (in press).



- R.T. Cooney, T.M. Willette, and S. Sharr. 1992 The effect of climate on Pacific salmon production in the northern Gulf of Alaska: examining the details of a natural experiment. In: Proceedings of the International Symposium on Climate Change and Northern Fish Populations, Can. Spec. Publ. Fish. Aquat. Sci.
- Willette, T.M. and R.T. Cooney. 1991 An empirical orthogonal functions analysis of sea surface temperature anomalies in the North Pacific Ocean and cross-correlations with pink salmon (*Oncorhynchus gorbuscha*) returns to southern Alaska. In: Proceedings of the 1991 Pink and Chum Salmon Workshop.
- Eggers, D.M., L.R. Peltz, B.G. Bue, and T.M. Willette. 1991. Trends in the abundance of hatchery and wild stocks of pink salmon in Cook Inlet, Prince William Sound, and Kodiak, Alaska. In: Proceedings of the International Symposium on the Biological Interactions of Enhanced Salmonids, Can. Spec. Publ. Fish. Aquat. Sci.

**B. Andrew Craig, Fisheries Biologist I**

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Commercial Fisheries Management and Development Division  
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(907)424-3212

PROJECT RESPONSIBILITIES: Design and supervision of embryo surveys, analysis, report writing

EDUCATION: 1990 Bachelor of Science, Fisheries Science, Cornell University

PROFESSIONAL EXPERIENCE:

1992 - present Fisheries Biologist I, Cordova, Commercial Fisheries Management and Development, ADFG

1991 - 1992 Fish and Wildlife Technician, Commercial Fisheries Division, ADFG

SELECTED PUBLICATIONS:

Craig, A.K., S. Sharr, and S.D. Moffitt. 1995. A compilation of historical preemergent fry and egg deposition survey data from Prince William Sound, 1961-1995. Alaska Department of Fish and Game, Division of Commercial Fisheries, Regional Information Report No. 2A95-49, Anchorage.

Bue, B.G., S. Sharr, S.D. Moffitt, and A.K. Craig. 1996. Effects of the *Exxon Valdez* oil spill on pink salmon embryos and preemergent fry. Pages 619-627 in S.D. Rice, R.B. Spies, D.A. Wolfe, and B.A. Wright, editors. Proceedings of the *Exxon Valdez* oil spill symposium. American Fisheries Society Symposium 18.

Sharr, S., B. G. Bue, S. D. Moffitt and A. K. Craig. 1994a. Injury to salmon eggs and preemergent fry in Prince William Sound - F/S 2. State/Federal Natural Resources Damage Assessment Final Report. *Exxon Valdez* Trustee Council, Anchorage, Alaska.

Sharr, S., J. E. Seeb, B. G. Bue, S. D. Moffitt, A. K. Craig and G. D. Miller. 1994b. Injury to salmon eggs and preemergent fry in Prince William Sound - R60C. State/Federal Natural Resources Restoration Final Report. *Exxon Valdez* Trustee Council, Anchorage, Alaska.

Sharr, S., J. E. Seeb, B. G. Bue, S. D. Moffitt, A. K. Craig and G. D. Miller. 1994c. Injury to salmon eggs and preemergent fry in Prince William Sound - 93003. State/Federal Natural Resources Restoration Final Report. *Exxon Valdez* Trustee Council, Anchorage, Alaska.

**C. David Evans, Biometrician I**

Commercial Fisheries Management and Development

Alaska Department of Fish and Game

Anchorage, Alaska 99518-1599

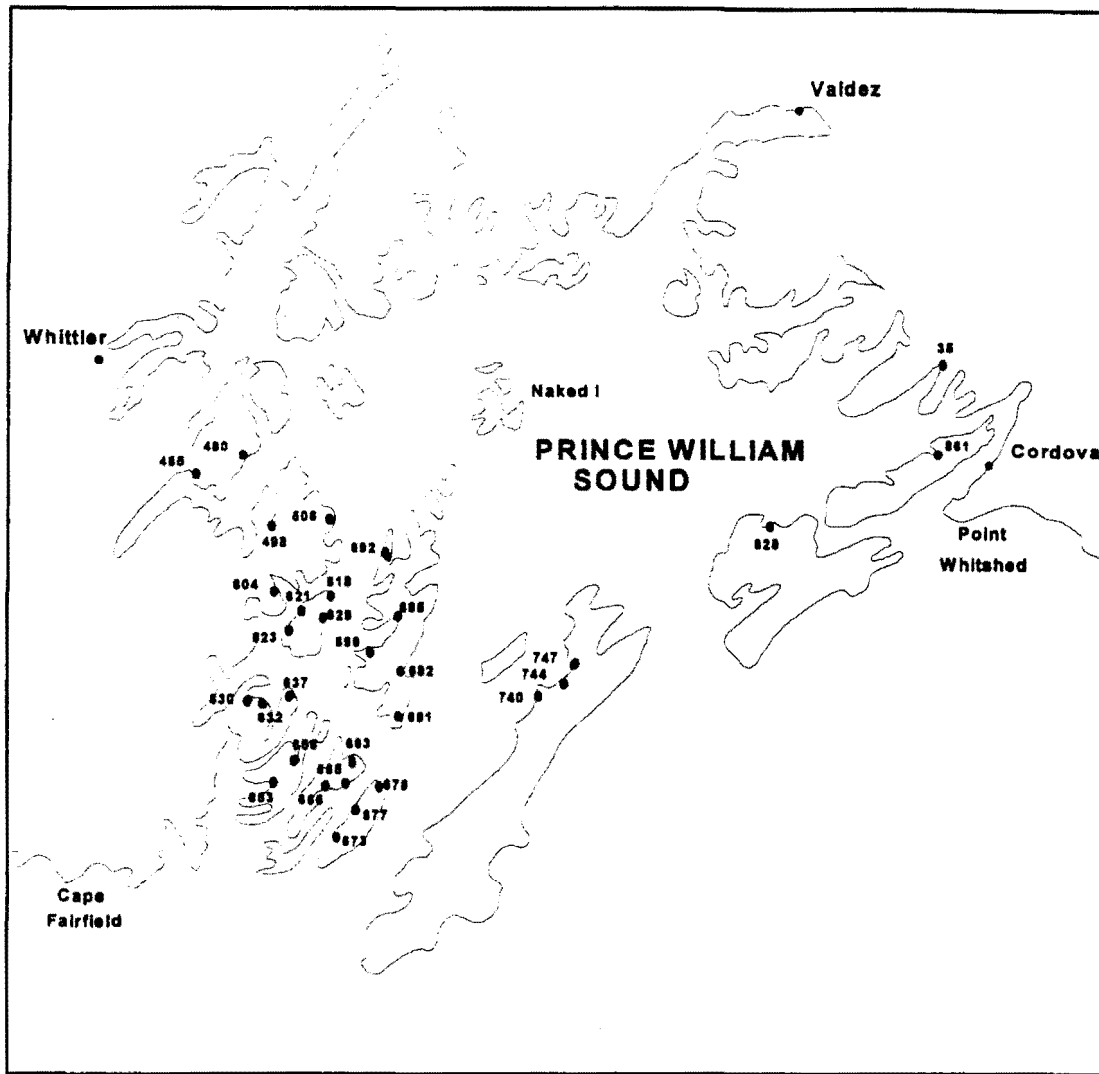
(907) 267-2176 DavidE%fishgame@state.ak.us

PROJECT RESPONSIBILITIES: Study design and analysis

EDUCATION: B.S., Soil Science, University of Nottingham (U.K.)  
M.S., Soil Science, University of Guelph (Ontario, Canada)  
M.S., Statistics, Oregon State University  
Ph.D., Soil Science, University of Guelph (Ontario, Canada)

PROFESSIONAL EXPERIENCE:

1991-present Biometrician I, CFMD, Alaska Dept. Fish and Game



**Figure 1.** Location of streams to be sampled for embryo deposition.

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# 1997 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1996 - September 30, 1997

Budget Category:	Authorized FFY 1997	Proposed FFY 1998						
Personnel	\$137.30	\$97.3						
Travel	\$13.90	\$5.7						
Contractual	\$87.60	\$36.6						
Commodities	\$24.30	\$5.3						
Equipment	\$2.10	\$2.1	LONG RANGE FUNDING REQUIREMENTS					
Subtotal	\$265.20	\$147.0	Estimated FFY 1999	Estimated FFY 2000	Estimated FFY 2001	Estimated FFY 2002	Estimated FFY 2003	Estimated FFY 2004
General Administration	\$26.7	\$17.2						
Project Total	\$291.9	\$164.2	\$58.7	\$0.0	\$0.0	\$0.0	\$0.0	\$0.0
Full-time Equivalents (FTE)		1.8						
Dollar amounts are shown in thousands of dollars.								
Other Resources								
Comments:								
Estimated costs for 'Field Monitoring' in FY 98.								
Costs for FY98 and beyond are estimated assuming that field monitoring will continue to show no difference in embryo mortality between oiled and non-oiled sites for two additional years. FY99 costs are for close out of field monitoring only.								

1998

Project Number: 98191A  
 Project Title: Investigating and Monitoring Oil Related Egg and Alevin Mortalities  
 Agency: AK Dept. of Fish & Game

FORM 3A  
 AGENCY  
 PROJECT  
 DETAIL

**1997 EXXON VALDEZ TRUST COUNCIL PROJECT BUDGET**

October 1, 1996 - September 30, 1997

Personnel Costs:			GS/Range/ Step	Months Budgeted	Monthly Costs	Overtime	Proposed FFY 1996	
PM	Name	Position Description						
	M. Willette	Fishery Biologist III	18F	2.0	6,309	0	12.6	
	A. Craig	Fishery Biologist I	14B	7.0	4,270	1,300	31.2	
	Vacant	6 - Fish and Wildlife Technician II & III	11A	7.0	3,992	1,000	28.9	
	D. Evans	Biometrician I	17E	4.0	5,279	0	21.1	
	P. Trautman	Field Office Assistant	11A	1.0	3,509	0	3.5	
Subtotal				21.0	23,359	2,300		
Those costs associated with program management should be indicated by placement of an *.							Personnel Total	\$97.3
Travel Costs:			Ticket Price	Round Trips	Total Days	Daily Per Diem	Proposed FFY 1996	
PM	Description							
	Attend biometrics consultation and EVOS mtg.		200	4	13	95	2.0	
	Attend pink and chum workshop		500	3	6	95	2.1	
	Attend state AFS meeting		400	3	4	95	1.6	
Those costs associated with program management should be indicated by placement of an *.							Travel Total	\$5.7

**1998**

Project Number: 98191A  
 Project Title: Investigating and Monitoring Oil Related Egg and Alevin Mortalities  
 Agency: AK Dept. of Fish & Game

FORM 3B  
 Personnel  
 & Travel  
 DETAIL



# 1997 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1996 - September 30, 1997

<b>Contractual Costs:</b>		Proposed
Description		FFY 1996
Vessel charter for Fall Embryo Sampling (R/V Montague @ \$1.2K/day for 25 days)		32.5
Air charter for Fall Embryo sampling (8 hours @ \$0.25/hour)		2.0
D.O.T. vehicle rental (2 months @ \$0.3/month)		0.6
Outboard maintenance(\$0.5)		0.5
Publication Costs		1.0
When a non-trustee organization is used, the form 4A is required.		
<b>Contractual Total</b>		<b>\$36.6</b>
<b>Commodities Costs:</b>		Proposed
Description		FFY 1996
Data processing supplies		1.5
Field sampling supplies (\$2.8)		2.8
Office Supplies		1.0
<b>Commodities Total</b>		<b>\$5.3</b>

**1998**

Project Number: 98191A  
 Project Title: Investigating and Monitoring Oil Related Egg and Alevin Mortalities  
 Agency: AK Dept. of Fish & Game

FORM 3B  
 Contractual &  
 Commodities  
 DETAIL

# 1997 EXXON VALDEZ TRUSTEES COUNCIL PROJECT BUDGET

October 1, 1996 - September 30, 1997

New Equipment Purchases:		Number of Units	Unit Price	Proposed FFY 1996
Description				
	Fry pump for field monitoring (Component A)			0.5
	Replacement outboard motor (25 hp) for field sampling			1.6
				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 a <b>New Equipment Total</b>				\$2.1
Existing Equipment Usage:		Number of Units	Inventory Agency	
Description				
	Hydraulic fry pumps	4	ADFG	

1998

Project Number: 98191A  
 Project Title: Investigating and Monitoring Oil Related Egg and Alevin Mortalities  
 Agency: AK Dept. of Fish & Game

FORM 3B  
 Equipment  
 DETAIL



## **Pink Salmon Spawning Habitat Recovery**

Project Number: 98194

Restoration Category:

Proposer: Michael L. Murphy and Stanley D. Rice  
NMFS, Auke Bay Laboratory  
ABL Program Manager: Dr. Stan Rice  
NOAA Program Manager: Bruce Wright

Lead Trustee Agency: NOAA

Cooperating Agencies:

Alaska SeaLife Center:

Duration: 2nd year, 1.5 year project

Cost FY 98: \$53.2K

Geographic Area: Prince William Sound (field work completed)

Injured Resource/Service: Pink salmon

**RECEIVED**  
APR 15 1997  
EXXON VALDEZ OIL SPILL  
TRUSTEE COUNCIL

### **ABSTRACT**

This proposal requests funds to close out Project 194 (Pink Salmon Spawning Habitat Recovery), allowing publication of results and participation at the 1998 Restoration Workshop. Project 97194 examined the level of oil contamination in pink salmon streams in 1989-90 and 1995 by analyzing sediment samples collected in 1989-90 by ADFG (Oil Spill Response) and similar samples collected in 1995 by the Auke Bay Laboratory. Approximately 500 samples from 200 streams were analyzed by the Auke Bay Laboratory in 1997. Results will help to complete the understanding of the injury to pink salmon by documenting the initial exposure level and subsequent habitat recovery. The requested funds would allow this project to fully analyze the data, prepare a manuscript for peer-reviewed publication, and participate at the 1998 Restoration Workshop.

## INTRODUCTION

The *Exxon Valdez* oil spill caused increased mortality and possible long-term genetic damage in pink salmon (*Oncorhynchus gorbuscha*) eggs and embryos that incubated in oiled intertidal sections of freshwater streams. In Fiscal Year 1997, Project 97194 (Pink Salmon Spawning Habitat Recovery) was funded to document initial levels of oil exposure and subsequent recovery of pink salmon spawning habitat. Results were meant to complement and help interpret other Trustee studies of oil-related embryo mortality in pink salmon.

To document initial exposure levels, the Auke Bay Laboratory (ABL) is analyzing approximately 400 sediment samples taken by the Alaska Department of Fish and Game (ADFG) from nearly 200 pink salmon streams in 1989-90. An additional 97 samples were collected by the ABL in 1995 from 12 of ADFG's sites (11 oiled, 1 non-oiled) and are being analyzed to determine habitat recovery. Most samples were taken from stream banks immediately adjacent to pink salmon spawning areas. Samples are being analyzed by ultraviolet fluorescence to measure total concentration of petroleum hydrocarbons, and representative samples are being analyzed by gas chromatography/mass spectroscopy to determine concentrations of individual polynuclear aromatic hydrocarbon (PAH) analytes to confirm the source of oil and determine its state of weathering.

Based on preliminary results, initial oil contamination of pink salmon spawning areas in 1989 varied widely, with mean oil concentration at streams ranging from 1  $\mu\text{g/g}$  to over 45,000  $\mu\text{g/g}$  of total oil hydrocarbons. In 1995, significant concentrations of petroleum hydrocarbons still remained in sediments at several streams. At the 11 oiled sites sampled in 1995, mean oil concentration ranged from 1  $\mu\text{g/g}$  to 240  $\mu\text{g/g}$ , and individual sediment samples were as high as 1,628  $\mu\text{g/g}$ . Analysis of the PAH "fingerprint" confirmed that the source of petroleum hydrocarbons was *Exxon Valdez* oil. A simple exponential decay model interpolating between mean oil concentration in 1989 and 1995 indicates that hydrocarbon levels probably still exceeded 1,000  $\mu\text{g/g}$  at many oiled sites until 1993. Preliminary GC/MS data from the 1995 samples indicates that some oil deposits continue to be relatively unweathered.

Initial levels of oil contamination at streams in Prince William Sound were sufficient to induce elevated mortality of pink salmon embryos incubating in stream gravel. Although impacts of oil on pink salmon eggs and embryos were evident after the *Exxon Valdez* oil spill, little information existed on the levels of oil contamination to which embryos were initially exposed. Some workers (Heintz and Weidmer 1996) inferred that PAH levels in 1990 were high enough to cause metabolic and mutagenic effects, but other workers (Brannon et al. 1996) disagreed, suggesting that PAH levels were too low to cause ill effects. Results from Project 97194 will now allow this question to be answered.

Although many streams had apparently recovered by 1995, some still had significant levels of oil that could cause impaired reproduction in pink salmon (Project 97194). Such residual oil levels could explain the persistent elevated embryo mortality observed in pink salmon from oiled streams in Prince William Sound. Sharr et al. (1994) suggested that increased mortality in embryos from adults taken from oiled streams and spawned at the AFK Hatchery was due to genetic damage. Results from Project 97194, however, indicate that embryo mortality could

have been elevated because the adults had been exposed to residual oil when they were embryos incubating in the oiled streams. Thus, the subsequent embryo mortality observed by Sharr et al. (1994) could have been a toxicological effect, rather than a genetic effect.

This proposal requests funds to close out Project 97194, allowing full analysis of results and their implications, publication of results in a peer-review journal, and participation at the 1998 Restoration Workshop. The funds provided for Project 97194 in Fiscal Year 1997 included only funds needed to analyze existing samples. The funds requested for Fiscal Year 1998 are to support the principal investigators during the close out of Project 97194, ensure that results are adequately analyzed, produce a final report, and produce a peer-reviewed literature publication.

## **NEED FOR THE PROJECT**

### **A. Statement of Problem**

Pink salmon embryos that incubated in intertidal sections of streams contaminated by the *Exxon Valdez* oil spill continued to show poor survival compared to those from non-oiled streams until 1994. The cause of the reduced survival is thought to be genetic damage from the initial acute exposure after the spill (Sharr et al. 1994). The mortality could also be due to continuing oil exposure from persistent oil deposits that seep toxic compounds into salmon spawning areas. Hydrocarbons carried by water percolating through the beach during ebb tide could become metabolically sequestered by incubating embryos, causing sufficient damage to impair reproduction when these exposed fish return to spawn as adults. Preliminary data on oil concentrations in stream and beach sediments from Project 97194 indicate high but variable initial exposure levels and continued significant exposure at some sites in 1995.

Upon full analysis, results from this study should provide valuable information on 1) the range of initial exposure levels and subsequent recovery, 2) causes of variable exposure and recovery (e.g., a bay's exposure to winter storms), 3) state of weathering of residual oil, and 4) relationship of residual oil contamination to observed egg/embryo mortality in pink salmon.

### **B. Rationale/Link to Restoration**

The proposed project would close out Project 97194 and allow full dissemination of results in a publication and workshop. It would make the data on initial oil concentrations in stream and adjacent beach sediments and on their 1995 condition widely available. This project relates directly to the Oil Spill Restoration Plan objective to restore pink salmon populations to prespill abundance, health, and productivity.

### **C. Location**

All 1995 samples were from within Prince William Sound. Samples from 1989-1990 were from the entire oil spill impact area, including Prince William Sound, Kodiak, and the Alaska and Kenai Peninsulas.

## COMMUNITY INVOLVEMENT

As all field work has already been completed, only limited community involvement is envisioned for this project.

## PROJECT DESIGN

### A. Objectives

The major hypotheses of Project 97194 were 1) that initial oil concentrations after the *Exxon Valdez* oil spill were sufficient to cause long-term genetic damage in pink salmon, and 2) that residual oil from beached deposits continues to seep into salmon spawning areas, contributing to poor embryo survival. Specific objectives for Fiscal Year 1997 were to:

Objectives in FY97:

1. Measure oil in ADFG-collected stream gravels collected in 1989-90;
2. Measure oil in ABL-collected stream gravels collected in 1995;
3. Examine PAH profiles in 1989-90 and 1995 samples and compare to *Exxon Valdez* crude for confirmation of oil source;
4. Prepare a report on the stream gravel concentrations, rate of recovery, and need and potential for restoration; and

All of the above objectives will be completed in the first year of Project 97194. The close-out portion of Project 194 would address the following additional objectives:

Objectives in FY98:

1. Analyze the data more fully to 1) determine causes of variability in initial concentration and subsequent recovery; 2) evaluate variation in state of weathering of the 1995 samples from the GC/MS data; 3) determine whether a relationship exists between PAH concentration in stream sediments and mussels (Project 090) from the same area; and 4) examine the relationship between oil concentrations in this study with Sharr et al.'s (1994) results on egg/embryo mortality from the same streams.
2. Prepare a manuscript for publication in a peer-reviewed journal describing the major results of Project 97194 and relating these results to other Trustee-sponsored studies of embryo mortality in pink salmon. It would discuss restoration strategies for pink salmon spawning areas and interpret previous Trustee-sponsored studies of embryo mortality in pink salmon.
3. Participate at the 1998 Restoration Workshop.

### B. Methods

After completing the final report for Project 97194, the principal investigators would test five hypotheses related to the FY98 objectives:

H1: Initial oil concentration and subsequent recovery are determined by a bay's orientation to prevailing currents and relative protection from storms. To test this hypothesis, data on initial concentration would be correlated to measures of bay orientation and exposure determined from maps. Effects of these variables on subsequent recovery would be tested by correlating them with the 1995 concentrations in the 12 streams sampled in 1995.

H2: Beach cleanup efforts after the oil spill helped to speed habitat recovery. To test this hypothesis, ADFG's records of oil cleanup efforts will be summarized for the 1995 study sites and compared to observed residual oil concentrations to determine the effect, if any, of cleanup efforts on subsequent habitat recovery.

H3: The state of oil weathering is related to a bay's exposure to beach overturn during storms. To test this hypothesis, an index of weathering (Short and Heintz, in press) would be computed for the 1995 samples and compared to data on bay orientation and exposure, sample location relative to tide level, and other data from detailed maps made at the 1995 study sites.

H4: Oil concentration in stream sediments is correlated with oil concentration in mussels and visual estimates of oiling in adjacent areas. Data on PAH concentration in mussels in 1989 and 1995 would be obtained from the State/Federal Trustee Council Hydrocarbon Database (Project 290) and compared with oil concentration in stream sediments (Project 97194) to determine whether mussel beds and stream sediments show similar trends in initial contamination level and subsequent recovery. Oil concentrations from all studies (Trustee-sponsored and Exxon contractors) would be compared to visual estimates of oiling from the ADEC Shoreline Survey, and all data would be tabulated in useful format.

H5: Egg and embryo mortality observed by Sharr et al. (1994) is related to residual oil concentrations and state of oil weathering at their sites. To test this hypothesis, data on 1989-90 and 1995 oil concentrations and weathering index would be compared with observed egg/embryo mortality at the same sites to determine whether residual oil could explain observed elevated mortality.

### **C. Cooperating Agencies, Contracts, and Other Agency Assistance**

No contracts or other agency assistance are anticipated.

## **SCHEDULE**

### **A. Measurable Project Tasks for FY 98**

October 1997 - March 1998: Analyze data; prepare manuscript for publication.

December 1997: Collate data from all projects and compare with stream sediments.

January 1998: Participate at the 1998 Restoration Workshop



## **B. Project Milestones and Endpoints**

March 1998: Manuscript submitted to peer-review journal for publication.  
May 1 1998: Final report submitted.

## **C. Completion Date**

This project would be completed in Fiscal Year 1988.

## **PUBLICATIONS AND REPORTS**

A peer-reviewed publication would be submitted by March 1998. This manuscript would address initial oil concentrations in 1989-90, habitat recovery in 1995, effects of beach cleanup efforts on habitat recovery, and causes of variation in habitat recovery. The final report would be completed by May 1998. It would include tables of oil concentrations from sediments and mussels from adjacent areas correlated with visual estimates of oiling.

## **NORMAL AGENCY MANAGEMENT**

NOAA NMFS has statutory stewardship for all living marine resources; however, if the oil spill had not occurred NOAA would not be conducting this project. NOAA NMFS proposes to make a significant contribution (as stated in the proposed budget) to the operation of this project, making it truly cooperative.

## **COORDINATION AND INTEGRATION OF RESTORATION EFFORT**

This project would be coordinated with other projects conducted by ABL. Much coordination has already been achieved during sample acquisition through shared logistics and data and in developing objectives for this project. Data from this project would be directly relevant to ongoing Restoration projects dealing with recovery of pink salmon and oil-related embryo mortality.

## **PROPOSED PRINCIPAL INVESTIGATORS**

Michael L. Murphy and Stanley D. Rice  
NOAA NMFS  
Auke Bay Laboratory  
11305 Glacier Hwy  
Juneau, AK 99801  
Phone: (907) 789-6036  
Fax: (907) 789-6094  
E-mail: mike.murphy@noaa.gov

## PRINCIPAL INVESTIGATORS

Michael L. Murphy, a GS-12 Fisheries Research Biologist, received a BA in Zoology from University of Wisconsin, Madison, in 1974 and an MS in Fisheries Science from Oregon State University, Corvallis, in 1978. Mike has been employed at the Auke Bay Laboratory since 1981. His principal studies have included research on stream/riparian habitat issues, watershed and stream ecosystems, and ecology of salmonids. He has published more than 40 papers and syntheses related to these topics. Mike is a nationally recognized expert on the effects of timber harvest on anadromous fish habitat, and has served on numerous invited science panels, including the National Interagency Salvage Program Review, the Tongass Land Management Plan EIS, the Interior Columbia Basin Forest Plan EISs, and the Alaska Science/Technical Committee advising the Board of Forestry on adequacy of the Alaska Forest Practices and Resources Act. Mike presently leads the Anadromous Fish Habitat Task at the Auke Bay Laboratory.

Dr. Stanley D. Rice, A GM-14 Physiologist, received a BA (1966) and MA (1968) in Biology at Chico State University, and Ph.D. (1971) in Comparative Physiology at Kent State University. Employed at the Auke Bay Laboratory since 1971 as a research physiologist and task leader, Dr. Rice has been Habitat Program Manager since 1986. He has researched oil effects since 1971 and has published over 80 papers on the subject. Dr. Rice was the lead editor for the *Exxon Valdez* Symposium published in 1977. His studies have ranged from field to lab tests, behavioral to biochemical studies, and salmonids to invertebrates. Dr. Rice has conducted and managed cooperative projects since 1974, including the Auke Bay Laboratory's *Exxon Valdez* damage assessment studies. Activities since the oil spill include management of 10 damage assessment projects, establishment of chemistry lab and analyses, and establishment of hydrocarbon database management. Dr. Rice has provided reviews and critical input to principal investigators and managers in NOAA and other agencies to support agency decisions, and he has interacted closely with other agencies on logistics coordination, study design review, and data interpretation.

## LITERATURE CITED

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- Sharr, S., J. E. Seeb, B. Bue, A. Craig, and G. D. Miller. 1994. Injury to pink salmon eggs and pre-emergent fry in Prince William Sound -- Restoration Study 93003. ADFG Regional Information Report No. 2A94-48. ADFG, Division of Commercial Fisheries Management and Development, 333 Raspberry Rd. Anchorage, AK 99518-1599.
- Short, J. W., and R. A. Heintz. In press. Identification of *Exxon Valdez* Oil in Sediments and Tissues from Prince William Sound and the northwestern Gulf of Alaska based on PAH Weathering. Environmental Science Technology.

# 1998 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1997 - September 30, 1998

Page 1

Budget Category:	Authorized FFY 1997	Proposed FFY 1998						
Personnel	\$102.7	\$35.7						
Travel	\$2.9	\$2.9						
Contractual	\$0.0	\$6.0						
Commodities	\$17.3	\$2.8						
Equipment	\$0.0	\$0.0	LONG RANGE FUNDING REQUIREMENTS					
Subtotal	\$122.9	\$47.4	Estimated FFY 1999	Estimated FFY 2000	Estimated FFY 2001	Estimated FFY 2002		
General Administration	\$15.4	\$5.8						
Project Total	\$138.3	\$53.2	\$0.0	\$0.0	\$0.0	\$0.0		
Full-time Equivalents (FTE)		0.4						
Dollar amounts are shown in thousands of dollars.								
Other Resources	\$37.7	\$11.2						
Comments: Other Resources:  FFY1998: NOAA contribution of ABL Habitat Program Manager, J. Rice, 1 mo = \$11.6 K.  FFY 1997: NOAA contribution of ABL Habitat Program Manager, J. Rice, 2 mo = \$ 22.4K: Senior Chemist, J. Short, 1 mo = \$ 8.3K: Principal Investigator Mike Murphy 1 mo = \$ 7K (in addition to mo. covered in next section) for a total of \$ 37.7K.  FFY 1996: NOAA contribution estimated at \$55.0K. This includes obtaining oil spill response samples from ADFG, consultation to identify and verify that collection data was compatible with the Trustee database, and field work to collect comparable samples in PWS in 1995. This project was not funded by the Trustee Council in FY95 nor FY96.								

**1998**

Project Number: 98194  
Project Title: Pink Salmon Spawning Habitat Recovery  
Agency: National Oceanic & Atmospheric Administration

FORM 3A  
TRUSTEE  
AGENCY  
SUMMARY

Prepared: 4/11/97

**1998 EXXON VALDEZ TRUST COUNCIL PROJECT BUDGET**

October 1, 1997      September 30, 1998

Page 2

<b>Personnel Costs:</b>		<b>GS/Range/ Step</b>	<b>Months Budgeted</b>	<b>Monthly Costs</b>	<b>Overtime</b>	<b>Proposed FFY 1998</b>
<b>Name</b>	<b>Position Description</b>					
M Murphy	Fishery Research Biologist	12/4	3.5	7.0		24.5
J Rice	Habitat Program Manager		1.0	11.2		11.2
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
<b>Subtotal</b>			4.5	18.2	0.0	
<b>Personnel Total</b>						<b>\$35.7</b>
<b>Travel Costs:</b>		<b>Ticket Price</b>	<b>Round Trips</b>	<b>Total Days</b>	<b>Daily Per Diem</b>	<b>Proposed FFY 1998</b>
<b>Description</b>						
Anchorage, Workshop & other Planning Mtgs., 2		0.4	2	6	0.3	2.6
Car Rental and miscellaneous for above						0.3
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
<b>Travel Total</b>						<b>\$2.9</b>

**1998**

Project Number: 98194  
 Project Title: Pink Salmon Spawning Habitat Recovery  
 Agency: National Oceanic & Atmospheric Administration

FORM 3B  
 Personnel  
 & Travel  
 DETAIL

Prepared: 4/11/97

**1998 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET**

October 1, 1997 - September 30, 1998

Page 3

<b>Contractual Costs:</b>		Proposed
Description		FFY 1998
NOAA contract labor: 300 hr x \$20/hr		6.0
When a non-trustee organization is used, the form 4A is required.		
<b>Contractual Total</b>		\$6.0
<b>Commodities Costs:</b>		Proposed
Description		FFY 1998
Computer repairs, maintenance, software upgrades		1.0
Production of Reports		1.8
<b>Commodities Total</b>		\$2.8

**1998**

Project Number: 98194  
 Project Title: Pink Salmon Spawning Habitat Recovery  
 Agency: National Oceanic & Atmospheric Administration

**FORM 3B**  
 Contractual &  
 Commodities  
 DETAIL

[illegible]

## 1998

Project Number: 98194  
Project Title: Pink Salmon Spawning Habitat Recovery  
Agency: National Oceanic & Atmospheric Administration

FORM 3B  
Equipment  
DETAIL



## **Pristane Monitoring in Mussels**

Project Number: 98195

Restoration Category: Research and Monitoring

Proposer: Jeffrey W. Short and Patricia M. Harris  
NMFS, Auke Bay Lab  
ABL Program Manager: Dr. Stan Rice  
NOAA Program Manager: Bruce Wright

Lead Trustee Agency: NOAA

Cooperating Agencies:

Alaska Sea Life Center:

Duration: 3 rd year, 5 year project

Cost FY98: \$115,000

Cost FY99: \$115,000

Cost FY00: \$ 75,000

Geographic Area: Prince William Sound

Injured Resource/Service: Pink Salmon, Pacific Herring

**RECEIVED**  
APR 15 1997  
**EXXON VALDEZ OIL SPILL  
TRUSTEE COUNCIL**

### **ABSTRACT**

This project will continue to monitor pristane in mussels as an indirect index of potential year-class strength for pink salmon and herring and to identify critical juvenile pink salmon and herring marine habitat in Prince William Sound (PWS).



## INTRODUCTION

Pristane is a hydrocarbon biosynthesized from chlorophyll by herbivorous copepods in the genera *Calanus* and *Neocalanus*. These copepods are the only proven modern marine source of pristane (Avigan & Blumer, 1968, J. Lipid Res. **9**:350:), and they typically contain concentrations that approach 1% dry weight (i.e. 10,000,000 ppb). As a branched alkane, pristane is highly lipophilic and resistant to metabolic degradation, which suggests that it may be a useful "tracer" molecule that would quantitatively label fats in predators of these copepods (Blumer *et al.*, 1964, Helgo. Wiss. Meeres. **10**:187). The low detection limit (about 100 ppb) of the inexpensive analytical method further suggests the utility of pristane as a natural indicator of energy flow from these copepods to higher trophic levels.

Pristane concentrations that range to 70,000 ppb (dry weight) are found in filter feeding organisms such as mussels and some clams in PWS during spring. Experiments conducted at the Auke Bay Laboratory in 1995 and 1996, confirm that pristane can be accumulated by mussels through ingestion of fecal material produced by predators of *Neocalanus*, e.g. juvenile pink salmon. In 1996, pristane concentrations in mussels at 3 sites near the W. H. Norenberg Hatchery on Esther Island increased dramatically within 2 to 6 days after the release of pink salmon fry from the hatchery. Released fry were observed feeding on *Neocalanus spp.* and defecating over mussels that were subsequently sampled, thereby confirming the rapid and efficient transfer of pristane from copepods to the mussels through salmon. Pristane concentrations in PWS mussels therefore reflect the timing and concurrent abundance of *Neocalanus spp.* and their predators in seawater adjacent to sampled mussels.

These results confirm that analysis of pristane in mussels may be used to investigate the PWS marine ecosystem. A regular monitoring of pristane in mussels may provide a quantitative basis for comparing inter-annual energy flow through *Neocalanus spp.* to commercially important predators such as herring and pink salmon. This may provide a relatively inexpensive indicator of survival through the early juvenile stages for these species. In addition, the monitoring program could identify locations where this flow is consistently high, i.e. essential fish habitats for these species. These approaches may clarify some of the important natural factors that affect recruitment of juvenile salmon and herring, which is necessary for determining the restoration of these resources. These areas of high energy flow would also be important to the many predators of the juvenile herring and salmon, including some of the marine bird species that have been identified as not fully recovered from the effects of EVOS.

Analysis of data collected during the pilot-study phase of this project (1994 and 1995) and under 96195 supports these conclusions:

Pristane concentrations throughout PWS remained low (generally below 300 ng/g dry weight) from September until mid-March when they began to increase. By the beginning of April, pristane concentrations had more than tripled in mussels at Knight Island Passage sites. These increases appeared to radiate over a wider area by late April, and by mid-May, pristane concentrations tripled again throughout most of PWS. From the mid-May peak, concentrations then gradually declined to the end of July, reflecting the descent of pristane producing copepods to overwintering depths below the near shore food web.

Mussels at sites adjacent to the deep marine trench (depths exceeding 300 m) in Knight Island Passage and adjoining, Wells Passage, Port Wells, and Port Nellie Juan had consistently high pristane concentrations in 1994, 1995, and 1996 relative to mussels at other sites, probably due to the overwintering of *Neocalanus* in the trenches. Despite inter-annual differences in the pristane-productivity index at some sites (see methods section), the productivity-index for the sound as a whole was similar in 1995 and 1996. In 1994, however, this index was somewhat lower, but this may be an artifact due to lower sampling frequency.

## **NEED FOR THE PROJECT**

### **A. Statement of Problem**

Determination of the causes of the dramatic declines in populations of pink salmon, herring and fish-eating seabirds following the *Exxon Valdez* oil spill requires an assessment of the natural factors that effect recruitment and survival of these species, because any negative effects of the spill may be confounded by these natural factors. In addition, natural factors impose constraints on the recovery potential of these species. Pink salmon have been identified as recovering; herring, pigeon guillemots, cormorants, and marbled murrelets are identified as not recovered (Invitation for Restoration Proposals 1998). If population declines of these species are the result of changes in the basic ecology of Prince William Sound due to natural phenomena (e.g. El Nino), then recovery of these populations to pre-spill levels may not be possible, and the criteria for recovery must recognize these changes.

### **B. Rationale**

The proposed project will continue to provide information that may be used to evaluate the effect of natural constraints on the recovery of Prince William Sound pink salmon and herring populations and secondarily, on fish-eating marine birds. Annual monitoring of pristane concentrations in mussels will permit an indirect evaluation of the effects of juvenile survival on recruitment. In addition, the monitoring will identify important near shore nursery areas for pink salmon and herring, the conservation of which may promote their recovery.

### **C. Location**

Mussel samples will be collected in Prince William Sound and will be analyzed for pristane

concentrations at the Auke Bay Laboratory, Juneau, Alaska. The identification of important productive areas in PWS and inter-annual productivity data will be useful to local fishery managers. Educational materials and the brochure will be most appropriate for residents and students of Prince William Sound, but will also be available for others.

## **COMMUNITY INVOLVEMENT**

We will continue to involve Prince William Sound residents in this project to share knowledge and interest in PWS ecosystems and to reduce sampling costs. Since 1994, the Prince William Sound Aquaculture Association has collected mussels near their 4 hatcheries at the appropriate times and stored them until the end of the season for pick-up. This year students with Youth Area Watch (Project 98210) and independent students will again be collecting mussels near their hometowns, Tatitlek, Whittier, Chenega, Kenny Cove, Valdez, Cordova, and Seward, and may be assisting with collections at other sites. We will provide materials for each participating school that explains the rationale of the project, and compares specific results for each school with the results for the whole effort. The underlying biology of this project gets to the heart of how the sound turns sunlight into fish, which we believe can provide a very useful local teaching resource. Youth Area Watch students will also continue to participate in a 1 day workshop at Auke Bay Laboratory on laboratory analysis techniques for pristane in mussels. A color brochure describing the project and reporting results will be updated to include 1997 data and will be available for volunteer collectors and others who are interested.

## **PROJECT DESIGN**

### **A. Objectives**

In 1998 this project has 2 objectives:

1. Measure pristane concentrations in mussels collected biweekly during spring from 36 stations in Prince William Sound to evaluate inter-annual variability of energy conversion from *Neocalanus* copepods to their near shore, shallow sea-depth predators (FY98 - FY00).
2. Determine the existence and location of regions inside Prince William Sound where the energy conversion of objective 1 is consistently above average, and synthesize these data over time and geographic location each succeeding project year (FY98 - FY00).

### **B. Methods**

Project objectives will be addressed by determining the seasonal variability of pristane concentrations in mussels (*Mytilus trossulus*) from 36 sites in Prince William Sound. Mussels will be collected biweekly, beginning in mid-March through June 1, then July 1 and August 1 for

a total of 9 collection periods and 324 mussel samples. The collection frequency is initially higher to more accurately establish the onset of the initial rise of pristane concentrations in the mussels, which is correlated with the zooplankton bloom and may vary from year to year. Collected mussels will be stored frozen and analyzed for whole-body pristane concentration.

Mussels (20) will be collected from selected mussel beds and placed into a plastic bag together with collection documentation (i.e. date, time, location, collector). Selected mussels will ideally be in the length range 20 - 45 mm. Mussels are collected along a transect parallel with the shoreline; 1 mussel is collected every consecutive meter. Previous results archived in the *Exxon Valdez* restoration database for hydrocarbons indicates that pristane concentrations in mussels collected in this way are representative of entire mussel beds.

Pristane concentrations in mussels will be analyzed statistically using least-significant difference (LSD) criteria based on an extensive sampling of the error distribution for these measurements. An error distribution for log-transformed pristane concentrations in mussels can be generated from 178 triplicate and 79 duplicate samples analyzed for the Exxon Valdez oil spill, which are contained in the *Exxon Valdez* Oil Spill of 1989: State/Federal Trustee Council Hydrocarbon Database 1989 - 1995 (EVTHD). These replicated samples were collected and analyzed by the same methods, and they all contained pristane concentrations above method detection limits. The variances of these replicates are homoscedastic after log transformation, so a distribution for differences of two random samples of the error distribution can be generated by Monte Carlo simulation. Based on this distribution of differences, the LSD at an  $\alpha = 0.05$  type I error rate is about 1.015, which corresponds to a ratio of about 2.75 for un-transformed data. Thus, mussels from two different samples are judged significantly different if the ratio of the larger pristane concentration to the smaller is more than 2.75. The power of this test to detect an actual increase of 3 is about 58%, again derived from Monte Carlo simulation of the error distribution. Since pristane concentrations in mussels typically increase by factors of greater than 10 during the season, the power of the sampling design is more than adequate.

Propagation of errors for derived indexes indicates that 66% increases of the pristane accumulation index (PAI) are significant at the  $\alpha = 0.05$  type I error rate. The PAI represents the productivity of near-shore *Neocalanus* consumers in one sampling season. The PAI is calculated as the product of pristane concentration and sampling interval, and is an approximation of the integral of concentration and time at each station. The power of these criteria to detect an actual doubling of the PAI is about 80%, estimated by Monte Carlo simulation. The power to detect differences among years for the sum of the PAI's across stations is even greater, due to the larger number of measurements involved: increases of 22% are significant, and the power to detect such increases when they occur is about 50%.

Results from 1996 indicate that the response time of pristane increases in mussels that results from a pulse increase of near-shore *Neocalanus* predators is about 4 days, assuming *Neocalanus* is abundant. The sampling schedule is sufficient to identify the timing of such increases to within about 2 weeks. Although more precision is desirable, it would be prohibitively expensive, involving at minimum a doubling of project cost. Conversely, the sampling scheme is probably

adequate to relate pristane increases observed in mussels with significant biological events such as the timing of wild and hatchery salmon outmigrations.

Specific hypotheses that will be addressed in 1998 are whether interannual variability of the PAI occurs randomly among stations. This will be evaluated statistically by Mantels test.

The chemical analysis of pristane involves pentane extraction of macerated tissues, lipid removal with silica gel, and separation and measurement of pristane by gas chromatography equipped with a flame ionization detector. Pristane measurement will use the internal standard method, with deuterated hexadecane and deuterated eicosane added to the pentane initially as the internal standard. Pristane identification will be based on retention time relative to the internal standard. Quality control samples include method blanks, spiked method blanks, and reference sample analyzed with each batch of 20 samples to verify method accuracy, precision, and absence of laboratory introduced artifacts and interferences. Recovery of the internal standard will be determine by adding a second internal standard prior to instrumental analysis. Method detection limits will be assessed annually for the mussel tissue matrix, and these detection limits will be assumed for the other matrixes analyzed. Based on previous performance, we anticipate accuracy of  $\pm 15\%$  of National Institute of Science and Technology (NIST)-certified values for the spiked blank and reference samples, precision of 95% of reference samples within  $\pm 15\%$  of sample means, and laboratory artifacts below detection limits more than 99% of the time. This level of analytical performance will insure that variability due to sample analysis is negligible compared with variability among replicate mussel samples.

Percent moisture and percent lipid will also be determined in samples so that results may be analyzed on dry weight and lipid weight bases. Dry weights will be determined by heating samples at 60 C to constant final weight. Lipid proportions will be determined from weight loss due to dichloromethane extraction.

Because there is no other practical way of estimating energy conversion from *Neocalanus* to their near-shore predators over a broad geographic area such as PWS, there are no alternative methodologies to consider here.

### **C. Contracts and Other Agency Assistance**

There will be no contracts under this project.

## **SCHEDULE**

### **A. Measurable Project Tasks for FY98**

FY98:

Oct 1 - Jan 1:	Analyze 1997 hydrocarbon data; revise brochure
Jan 1 - Feb 1:	Prepare and present 1997 data at Restoration

	workshop.
Feb 1-Mar 15:	Plan logistics for FY98 field season.
Feb 1 - Apr 15:	Prepare annual 1997 report and report for public & high schools
Mar 15 - Aug 1:	Collect mussel samples.
Aug 1 - Sep 30:	Analyze samples for pristane.

## **B. Project Milestones and Endpoints**

Objectives 1 & 2 should be met by FY00, possibly sooner, depending on the results. The endpoints are completion of the statistical analyses described under Methods above.

## **C. Completion Date**

The monitoring element will be performed annually for 3 more years; FY 98 through FY 00.

## **PUBLICATIONS AND REPORTS**

This project requires consistent multi-year funding to be successful. Annual reports are therefore appropriate, but publication in a peer-reviewed journal is also anticipated. In FY98, there will be 4 consecutive years of consistent monitoring results available, which will support at least one professional paper to be completed that year. Annual reports will be submitted on April 15 of each year. Quarterly reports will be prepared on forms supplied by the Restoration Office.

## **NORMAL AGENCY MANAGEMENT**

Although NOAA NMFS has statutory stewardship for all living marine resources, NOAA is conducting this project only because the oil spill occurred and marine resources were injured. NOAA NMFS will, however, make a significant contribution (as stated in the proposed budget) to the operation of this project.

## **COORDINATION AND INTEGRATION OF RESTORATION EFFORT**

We are cooperating closely with Youth Area Watch (98210), which is providing us with samples and to whom we are providing training and educational materials. Data and results will be shared with other projects, especially with Sound Ecosystem Assessment (SEA 98320), Alaska Predator Ecosystem Experiment (APEX 98163) and related seabird projects as restoration studies mature and the ability to integrate results becomes more possible.

## **EXPLANATION OF CHANGES IN CONTINUING PROJECTS**

No changes are proposed.

## **PROPOSED PRINCIPAL INVESTIGATOR**

Jeffrey W. Short  
Auke Bay Laboratory, Alaska Fisheries Science Center  
National Marine Fisheries Service, NOAA  
11305 Glacier Highway, Juneau, Alaska 99801-8626  
Phone: (907) 789-6065  
FAX: (907) 789-6094  
e-mail: jeff.short@noaa.gov

## **PERSONNEL**

Principal Investigator:

Jeffrey W. Short

Education:

BS, 1972, University of California, Riverside (Biochemistry & Philosophy)

MS, 1982, University of California, Santa Cruz (Physical Chemistry)

Relevant Experience:

1989- Present: Established and managed the hydrocarbon analysis facility at ABL to analyze hydrocarbon samples generated by the *Exxon Valdez* NRDA effort (about 20% of these samples were analyzed at ABL).

1989 - 1992: Principal Investigator, Exxon Valdez project Air/Water #3: Determination of petroleum hydrocarbons in seawater by direct chemical analysis and through the use of caged mussels deployed along the path of the oil spill.

1991 - 1996: Principal Investigator, Exxon Valdez project Subtidal #8: Development of

Prepared 3/97

Project 98195

computer-based statistical methods for global examination of sediment and mussel hydrocarbon data produced for the Exxon Valdez NRDA effort for systematic bias, and for identification of probable sources of hydrocarbons. In addition, this project produced both hard-copy and computer display maps of all the sediment and mussel hydrocarbon data.

1994 - 1995: Initiated data analysis and pilot projects that established the role of pristane in Prince William Sound.

1996-1997 Principal Investigator 96195 and 97195

#### Other Key Personnel:

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

#### Relevant Experience:

1989-1992: Co-principal investigator of NRDA study Subtidal 3, was responsible for field logistics and sample collection and assisted in data analysis and report preparation; also assisted other NRDA projects in field collections.

1992 -1996: participated in study design, field work, proposal preparation, data analysis, and report preparation for mussel bed monitoring and restoration (R103-96090).

1994-1997 Participated in logistic planning, sampling, and community involvement coordination for the pilot pristane project ,96195, and 97195.

Relevant publications: Co-author of final reports for NRDA study Subtidal 3 and several publications pertaining to distribution of *Exxon Valdez* oil in mussels and underlying sediments. Several public presentations of oil-related scientific research.

Responsibilities: Coordinate sample collection logistics and collect mussel samples; data analysis; report and proposal preparation; and preparation of science educational materials, posters, and reports.



# 1998 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1997 - September 30, 1998

Budget Category:	Authorized FFY 1997	Proposed FFY 1998						
Personnel	\$56.6	\$28.5						
Travel	\$43.2	\$42.6						
Contractual	\$0.2	\$30.5						
Commodities	\$6.8	\$6.9						
Equipment	\$0.0	\$0.0						
Subtotal	\$106.8	\$108.5	LONG RANGE FUNDING REQUIREMENTS					
General Administration	\$8.5	\$6.4	Estimated FFY 1999	Estimated FFY 2000	Estimated FFY 2001	Estimated FFY 2002	Estimated FFY 2002	
Project Total	\$115.3	\$114.9	\$115.0	\$115.0	\$75.0	\$0.0		
Full-time Equivalents (FTE)	0.8	0.5						
Dollar amounts are shown in thousands of dollars.								
Other Resources		\$61.2						
Comments:								
<p>NOAA contribution towards this project:</p> <p>Habitat Program Manager, S. Rice, 1 mo. @ \$9.6K</p> <p>Senior Research Chemist, Principal Investigator, J. Short, 6 mo. @ \$6.8K = \$40.8</p> <p>Total contribution = \$50.4K</p>								

1998

Project Number: 98195  
 Project Title: Pristine Monitoring in Mussels  
 Agency: National Oceanic and Atmospheric Administration

FORM 3A  
 TRUSTEE  
 AGENCY  
 SUMMARY

**1998 EXXON VALDEZ TRUSTE COUNCIL PROJECT BUDGET**

October 1, 1997 - September 30, 1998

<b>Personnel Costs:</b>		<b>GS/Range/ Step</b>	<b>Months Budgeted</b>	<b>Monthly Costs</b>	<b>Overtime</b>	<b>Proposed FFY 1997</b>
<b>Name</b>	<b>Position Description</b>					
P. Harris	Zoologist	9/6	3.5	4.2		14.7
L. Holland	Research Chemist	11/7	2.0	5.2		10.4
J. Short	Senior Research Chemist	13/0	0.5	6.8		3.4
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
<b>Subtotal</b>			6.0	16.2	0.0	
<b>Personnel Total</b>						<b>\$28.5</b>
<b>Travel Costs:</b>		<b>Ticket Price</b>	<b>Round Trips</b>	<b>Total Days</b>	<b>Daily Per Diem</b>	<b>Proposed FFY 1997</b>
<b>Description</b>						
Anchorage Workshop & technical review session		0.4	3	9	0.2	3.0
						0.0
Cordova & PWS to collect mussels. 9 trips						0.0
Alaska Airlines & food & lodging		0.4	9	30	0.2	9.6
						0.0
air charter (30 days flying averaging \$1K/day)		1.0	30			30.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
<b>Travel Total</b>						<b>\$42.6</b>

**1998**

Project Number: 98195  
 Project Title: Pristine Monitoring in Mussels  
 Agency: National Oceanic and Atmospheric Administration

FORM 3B  
 Personnel  
 & Travel  
 DETAIL

Prepared: 2 of 4

4/8/97

4/11/97

# 1998 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1997 - September 30, 1998

<b>Contractual Costs:</b>		Proposed
Description		FFY 1997
Transportation of samples		0.2
NOAA Contract Labor: 1515 hr x \$20/hr		30.3
NOAA considers air charters as travel costs & are listed under Travel		
When a non-trustee organization is used, the form 4A is required.		
<b>Contractual Total</b>		<b>\$30.5</b>
<b>Commodities Costs:</b>		Proposed
Description		FFY 1997
Chemicals, solvents for pristane analyses		2.0
Chemistry lab supplies (consummaables, glassware, equipment repairs)		2.5
Collecting gear and supplies (coolers, blue ice, plastic bags, film, etc.)		2.0
Project informational brochures for the public		0.4
<b>Commodities Total</b>		<b>\$6.9</b>

**1998**

Project Number: 98195  
 Project Title: Pristane Monitoring in Mussels  
 Agency: National Oceanic and Atmospheric Administration

FORM 3B  
 Contractual &  
 Commodities  
 DETAIL

ired:

3 of 4

4/8/97

1/11/97

**1998 EXXON VALDEZ TRUST      COUNCIL PROJECT BUDGET**

October 1, 1997 - September 30, 1998

<b>New Equipment Purchases:</b>		Number of Units	Unit Price	Proposed FFY 1997
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 purchases associated with replacement equipment should be indicated by placement of an R.		<b>New Equipment Total</b>		\$0.0
<b>Existing Equipment Usage:</b>		Number of Units	Inventory Agency	
Description				
camers		1	NOAA	
computer, NEC monitor		1	NOAA	
printer		1	NOAA	
color printer		1	NOAA	
VHS Radio		1	NOAA	
GPS unit		1	NOAA	
freezer		1	NOAA	
GC/MS		1	NOAA	

**1998**

Project Number: 98195  
 Project Title: Pristane Monitoring in Mussels  
 Agency: National Oceanic and Atmospheric Administration

**FORM 3B  
Equipment  
DETAIL**

Prepared: 4 of 4

4/8/97

4/11/97



## Genetic Structure of Prince William Sound Pink Salmon

Project Number: 98196

Restoration Category: Research and Monitoring

Proposer: Alaska Department of Fish and Game

Lead Trustee Agency: Alaska Department of Fish and Game

Cooperating Agencies: None

Alaska SeaLife Center: No

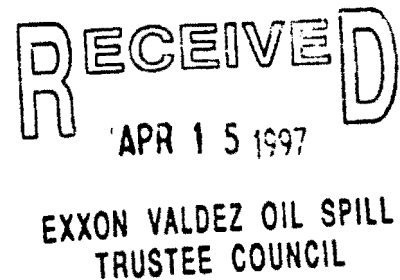
Duration: 5th year, 6-year project

Cost FY 98: \$130,200

Cost FY 99: \$50,000

Geographic Area: Prince William Sound

Injured Resource/Service: Pink Salmon



### ABSTRACT

Previous workers found that wild-stock pink salmon suffered both direct lethal and sublethal injuries as a result of the *Exxon Valdez* oil spill. An understanding of the population structure of pink salmon in Prince William Sound is essential to assess the impact of these injuries on a population basis and to devise and implement management strategies for sustained conservation. Results to date from this study suggest gene flow between pink salmon spawning aggregates can be restricted both spatially (regional and upstream-tidal) and temporally (early-late) within the Sound. This proposal increment covers the final year of laboratory analysis and the statistical analysis of year-three allozyme and mtDNA data.

## INTRODUCTION

In this continuing project we delineate the genetic structure of populations of wild pink salmon (*Oncorhynchus gorbuscha*) inhabiting Prince William Sound (PWS). We are testing for both temporal and geographical structuring among even- and odd-year races by examining genetic differences between early- and late-season spawners, upstream and tidal spawners, and among stream of spawning. This knowledge of genetic structure will be used in order to:

- A. Correctly interpret and apply the findings obtained from the proposed ecosystem analyses (98320) on a population basis.
- B. Provide genetic information needed for risk assessment and genetic monitoring of supplementation programs (e.g., proposed as a result of Trustee Council Projects R105, 95320 A-P, 95093, 96320 or 97320) to guide population-specific restoration and enhancement.
- C. Better direct harvest management decisions made for conservation purposes on a population-specific rather than species-specific basis. Our goal is to provide the basis for key management decisions by defining the genetic structure of representative populations from throughout PWS, measuring both within- and between-population diversity.

We propose to examine spawning aggregates from the even-year broodline and the odd-year broodline each for two years. Two years of analysis are needed in order to confirm stability of population structure across years.

To date the Trustee Council has funded collection of 34 odd- and 51 even-year putative populations for genetic analyses. A comprehensive suite of both nuclear (allozyme) and mitochondrial (mtDNA) markers is being screened. In 1994 and 1995 we contracted with Washington Department of Fish and Wildlife for the laboratory analysis of 32 even-year and 16 odd-year collections using allozymes. We conducted the mtDNA analyses and the data analyses. Results from the even-year class show significant differences between upstream and intertidal spawning aggregates within two streams; we also observed significant differences between southwest-Sound and east-Sound collections. The 1995 samples focused on early-late, and tidal-upstream comparisons. Preliminary results using allozyme data from these samples indicates that there are stream-to-stream differences and temporal (early-late) differences within two of the three streams examined, but we did not detect any tidal-upstream differences. The mtDNA data also showed stream-to-stream differences but did not suggest differences between early and late or upstream and tidal collections.

## NEED FOR THE PROJECT

### A. Statement of Problem

Historically, wild stocks produced approximately five-hundred-million pink salmon fry which

emerged from streams throughout PWS each year to migrate seaward. Adult returns of wild pink salmon averaged from 10 to 15 million fish annually. Unlike returns of adult hatchery fish, these returning wild-stock adults play a critical role in the total PWS ecosystem: they convey essential nutrients and minerals from the marine ecosystem to estuaries, freshwater streams, and terrestrial ecosystems. Both juveniles and adults are important sources of food for many fishes, birds, and mammals. Wild pink salmon also play a major role in the economy of PWS because of their contribution to commercial, sport, and subsistence fisheries in the area.

Wild-stock pink salmon suffered both direct lethal and sublethal injuries as a result of the *Exxon Valdez* oil spill (EVOS). Pink salmon embryos and alevins suffered increased mortality, diminished growth, and a high incidence of somatic cellular abnormalities as a result of spawning ground contamination and rearing in oiled areas. Elevated mortality of embryos in the oiled streams has continued through 1993, three generations after the oiling, suggesting that genetic damage may have occurred (Craig et al. 1996). In controlled incubation, oiled substrate resulted in increased mortality of pink salmon to the eyed stage (Marty et al. *In press*). Also, in 1989 the commercial harvest of pink salmon had to be shifted away from the hatchery and wild stocks in the oiled areas to target only the wild stocks in eastern PWS. This resulted in over-harvest and depletion of these stocks evidenced by general run failures of eastern PWS stocks of non-hatchery origin in 1991.

PWS is also the center of one of the State of Alaska's largest aquacultural industries. Alaska Department of Fish and Game (ADFG) has been grappling with management of the wild stocks in face of complicated hatchery/wild-stock interactions for nearly a decade. The EVOS-related damages to wild stocks, coupled with full-scale hatchery egg takes, has exacerbated wild-stock management concerns. The commercial fishing industry and the two aquaculture associations are facing serious financial challenges due to the alterations in management imposed as a result of declines in abundance of wild pink salmon.

## **B. Rationale/Link to Restoration**

It is essential to manage and restore the damaged pink salmon resources on a population basis in order to conserve between-population diversity. While "stock" is used by biologists as a convenient term designating fish that spawn at a certain time at a certain place, stocks may not be genetically distinct from each other; also, a stock may be composed of multiple genetically divergent groups. "Population" describes genetically distinct groups of fish which are the building blocks of species. Gene flow is restricted between populations (thus carbon flow is restricted--see related proposals in Trustee Council project 98320), and this resulting between-population diversity is responsible for many aspects of the fitness of the species. In the case of commercially harvested species like pink salmon, fitness is defined to include the peak productivity and long-term sustainability. Between-population diversity provides optimal production for species inhabiting diverse ecosystems such as PWS; highly diverse population mixes also provide a biological buffer to environmental change (droughts, floods, major earthquakes, and other routine events that occur in Alaskan ecosystems).

Understanding genetic structure of the wild stocks inhabiting PWS is critical to their



management and conservation. For example, managing on too fine a scale may adversely affect the fishing industry and waste management resources, while managing on too large a scale may result in loss of genetic adaptations and diversity in the wild pink salmon populations within PWS. Knowledge gained through this project is needed to correctly interpret and apply the findings obtained from the proposed ecosystem analyses on a population basis, more properly define the population-level nature of the damage documented in previous study of EVOS-damaged pink salmon, and otherwise guide the decision-making process in the management-oriented restoration of the EVOS-damaged pink salmon populations. The same knowledge of population structure will be used for genetic monitoring and risk assessment, required to evaluate any supplemental restoration programs. This monitoring and risk assessment is analogous to the process currently being conducted to evaluate supplemental restoration of damaged populations on the Columbia River by the Northwest Power Planning Council (Waples et al. 1991).

Even- and odd-year classes have independent population structures because of the rigid two-year life cycle of pink salmon. For example, climactic, tectonic or other such events (such as the 1964 earthquake or the 1989 oil spill) may affect the population structure of the either odd or even year classes while leaving the alternate year-class relatively unchanged. Therefore, we are examining the population structure of both even- and odd-year classes.

We are continuing to examine population structure by using both nuclear (using allozyme electrophoresis) and mtDNA approaches in this ongoing project. Both allozyme analysis and mtDNA analysis will be used to discriminate populations and describe population structure. Genetic studies using allozyme analysis have proven especially useful for the conservation and management of populations of pink salmon (e.g., Shaklee et al. 1991; White and Shaklee 1991); we are also expanding our pilot analysis using mtDNA analyses, as our preliminary data have shown potential usefulness for detecting restricted gene flow between groups of PWS spawners.

Allozyme analysis remains the preferred approach for study of population genetics of salmonids because of its power to resolve populations of many species in the tetraploid-derived family by assaying many nuclear loci rapidly and at low cost (Allendorf 1994). Additional advantages of allozymes in this study include the fact that a pre-spill allozyme data set exists for comparison, and also many laboratories cooperate on inter-institutional examinations of pink salmon using allozymes, providing a support structure including a wealth of compatible data for comparison among Pacific rim populations (e.g., Beacham et al. 1985, 1988; Shaklee et al. 1991; White and Shaklee 1991; Shaklee and Varnavskaya 1994).

The utility of mtDNA approaches varies with the organism under study for reasons such as high relative cost and slow relative throughput (Allendorf 1994) ; additionally, sometimes mtDNA data reveal less diversity than that detected through allozymes because mtDNA loci are absolutely linked, cannot recombine, and are maternally inherited as a single locus (Smouse et al. 1994). However, adjacent pink salmon populations tend to be closely related (Shaklee and Varnavskaya 1994), and our FY 95 haplotype data indicate an east-west-island and upstream-intertidal separation of populations within PWS. We believe that the complementary use of the two techniques should provide optimal resolution of the population structure for this study.

## **C. Location**

The field portion of this project will be conducted in PWS (based out of Cordova, Alaska); the allozyme and the mtDNA analysis, experimental matings and fish culture, and data analyses will be completed in Anchorage, Alaska. The laboratory and fish-rearing portions of the project will be moved to the Alaska SeaLife Center in Seward when that facility is available.

## **COMMUNITY INVOLVEMENT AND TRADITIONAL ECOLOGICAL KNOWLEDGE**

This project also had strong support from the Prince William Sound Aquaculture Corporation and the Cordova fishing community when it was first drafted in 1991. Wherever possible, local-hire will be used to fill field positions required for sampling or for routine laboratory positions. People from the communities in PWS will have an opportunity to participate in this project as employees of the ADFG which gives local residents priority in hiring for state employment. Local knowledge from years of pink salmon fishery management was used in defining the hypotheses for testing for genetic homogeneity. For example, bimodal distribution in run timing observed in pink salmon runs in some streams within PWS lead us to include temporal genetic isolation as one of the hypotheses to be tested. Results from this study have been and will continue to be disseminated to the local community through Regional Planning Team meetings.

## **PROJECT DESIGN**

### **A. Objectives**

Our objective is to test the null hypothesis of panmixia among of pink salmon collections in the EVOS-affected area of PWS. Our specific objectives are to test the following:

1. there are no genetic differences between upstream and intertidal pink salmon spawners within the same streams.
2. there are no genetic differences between pink salmon spawners from different streams within PWS.
3. there are no genetic differences between pink salmon spawners from different regions within PWS.
4. there are no genetic differences between pink salmon spawners with different run timings within the same streams.
5. there are no genetic differences between odd- and even-year pink salmon spawners.
6. inheritance of putative allozyme alleles and loci follows Mendelian ratios.

## B. Methods

### 1. Field Sampling

#### *Physiography of Prince William Sound*

Tissues for baseline genetic data will be collected from up to 100 individuals from each of 30 spawning aggregations of each year class. Sampling will be based on the physiography of PWS and will include areas uplifted and areas unaffected by the 1964 earthquake (Figure 1). Sampling locations will incorporate a broad geographical distribution within the Sound (Table 1) including three hatcheries (Solomon Gulch, Cannery Creek and Armin F. Koernig) and 27 spawning aggregates from wild-stock streams.

The overall sampling design was guided by needs outlined in numerous meetings with ADFG fisheries managers and regional management biologists. Sampling will be done to include at least one collection from each of the five major management districts designated for pink salmon (Southeast, East, North, Southwest, Montague; Figure 2). The Sound was historically divided into a total of nine districts for management and conservation purposes according to target species and other biological, geographical, and geological factors (Anonymous 1960; Randall et al. 1983; Rugolo 1984). Because of study objectives, four of these smaller districts were incorporated into the five major districts. Three of these (Unakwik, Coghill, and Eshamy) were originally delimited for sockeye salmon management and will be combined into the North (Unakwik and Coghill) and Southwest (Eshamy) districts for this study. The Northwest district, a small region originally delineated for chum salmon and pink salmon management, will be combined into the North district for this study because of its small size, geographic location, and run timing similarities of pink salmon inhabiting these two districts.

Sampling will be designed to include both early and late stocks and inter-tidal and upstream-spawning stocks. Because abundance of pink salmon varies annually, selection of spawning aggregations will be determined by field personnel who will be instructed to sample streams that maximize the ability to investigate temporal (between years and within years) and spatial (between streams and within streams) comparisons. Tissue samples from heart, liver, muscle, and vitreous humor from each individual will be immediately frozen on liquid nitrogen and stored in Anchorage at -80°C.

### 2. Laboratory Analysis

#### *Allozymes*

Genetic data will be collected using the techniques of allozyme electrophoresis on all samples (Utter et al. 1987; Seeb et al. 1987). A pre-spill data base of allozyme

frequencies from 12 loci exists for PWS pink salmon (Seeb and Wishard 1977) which facilitates analyses of potential changes of population structure and gene flow. An extensive allozyme screening was undertaken by Washington Department of Fish and Wildlife (WDFW), subcontractor on this project in 1994-95, to maximize the potential number of available gene markers for examination in this project. The 77 loci resolved (Table 2) are greater in number than those examined in any previous study (Beacham et al. 1988; Shaklee et al. 1991; Shaklee and Varnavskaya 1994).

Allozyme techniques will follow those of Harris and Hopkinson (1976), May et al. (1979), and Aebersold et al. (1987); nomenclature will follow the American Fisheries Society standard (Shaklee et al. 1990). Gels will be scored using the on-line scoring program developed by the ADFG Genetics Laboratory. This collection and management system provides extensive documentation of results and error checking capabilities; it also facilitates rapid collation, analysis, and reporting of genetic data in order to ensure rapid turnaround, complete documentation, and immediate availability of summary statistics.

*S-plus* analytical software (Mathsoft, Inc., Seattle WA) will be used to calculate allele frequency estimates, to test for conformation of genotype frequencies to Hardy-Weinberg expected frequencies using log-likelihood ratios, and calculate Nei's (1978) genetic distance and Cavalli-Sforza and Edwards (1967) genetic distance. This application will also be used to perform hierarchical analyses using log-likelihood (modified from Weir 1990) to determine if significant population substructuring exists among PWS pink salmon based on the following criteria: even versus odd-year, upstream versus intertidal spawning location, early versus late run, and geographic location of spawning. Sequential Bonferroni corrections (Rice 1989) will be used to adjust significance levels.

We will estimate genetic relationships by deriving UPGMA (Sneath and Sokal 1973) and neighbor-joining trees (Saitou and Nei 1987) with Cavalli-Sforza and Edwards (1967) genetic distance and a UPGMA tree with Nei's (1978) genetic distance. In addition multi-dimensional scaling (MDS, Lessa 1990) will be performed using Cavalli-Sforza and Edwards (1967) genetic distances. MDS is an ordination technique that plots genetic relationships in two dimensions so that the plotted distances between collections closely match the observed distances in multidimensional space. This technique provides a means to confirm expected structure and uncover unexpected structure by providing insight into structural demarcations. All calculations will be performed using functions in *S-Plus*.

Finally, all allozyme data will be merged into the state and federal inter-agency databases maintained by NMFS, ADFG, and WDFW.

#### *Mitochondrial DNA*

An initial screening with 20 restriction enzymes was done in 1995 to identify polymorphic sites in both even- and odd-year cohorts (Table 3). Samples for the

screening came from spawning aggregates from tidal collections from three geographically separated streams (Duck River, Swanson Creek, and Humpback Creek; Seeb et al. 1996). Six enzymes (*ApaI*, *BstU I*, *EcoR V*, *Hinf I*, *Rsa I*, *XbaI*) that detected polymorphisms in the *ND5/6* region were then used to screen the remaining even-year collections. We propose to continue the screening to include individuals from upstream and tidal collections (Table 1) because of the differences observed in haplotype frequencies between upstream and tidal collections thus far (Seeb et al. 1996). We will reevaluate the six-enzyme screen that is proposed for all remaining collections should new polymorphic sites be detected. A target sample size of 40 will be set for all collections.

DNA will be extracted using Puregene DNA isolation kits for animal tissues (Gentra Systems, Inc. P.O. Box 13159, Research Triangle, N.C. 27709-13159). This process includes: (1) a cell lysis solution to break down cell and nuclear membranes; (2) a Proteinase K digest to denature proteins; (3) an RNase treatment to digest RNA; (4) protein precipitation to remove Proteinase K, RNase, and denatured proteins; (5) isopropanol to precipitate DNA; (6) 70% ethanol to wash DNA; and finally (7) a hydration solution to rehydrate DNA.

After extraction, the DNA will be amplified using the polymerase chain reaction (PCR; Saiki et al. 1988; Kocher et al. 1989). Amplified DNA will be cut with about seven restriction enzymes found to detect haplotype polymorphisms (of the 20 screened ; Table 3) and electrophoresed on agarose gels. Fragments will be visualized under UV light, and a photographic record will be made of each gel.

Since genes which are encoded by the mitochondrial genome are inherited as a single unit (i.e., analogous to linked loci), the restriction sites detected for each enzyme, for all regions examined, will be pooled as composite haplotypes. The frequencies and distributions of these composite haplotypes will then be used to examine the structure of salmon populations.

Nucleotide ( $\pi$ ) and haplotype ( $h$ ) diversity measures (Nei 1987) will be calculated for all collections using the restriction enzyme analysis package (*REAP*; McElroy et al. 1992). These measures estimate the number of nucleotide substitutions per site between DNA sequences (i.e., sequence divergence) and the amount of DNA polymorphism within collections, respectively.

To test for heterogeneity among populations, Monte Carlo simulations with 10,000 replicates will be performed (Roff and Bentzen 1989) using the *REAP* analysis program. Independent tests will be performed to test for heterogeneity in a hierarchical manner following the levels identified in the log-likelihood analysis. However, unlike the log-likelihood analysis, the  $\chi^2$  values for individual tests are not summable. Monte Carlo tests will also be performed between the paired upstream and tidal collections, and among-region tests will be conducted by pooling collections within region. Significance levels will be adjusted using sequential Bonferroni techniques (Rice 1989).

An analysis of the distribution of molecular variance will be made using AMOVA (Excoffier et al. 1992) and utilizing a matrix of Euclidean distances between haplotypes. Pairwise Euclidean distances will be calculated as the total number of site changes between haplotypes. The AMOVA analysis incorporates distance between haplotypes in the calculation of haplotypic diversity at different hierarchical levels. Haplotype correlation measures are expressed as  $\Phi$ -statistics (Excoffier et al. 1992). Among regions,  $\Phi_{CT}$  is defined as the correlation of random haplotypes within a group of collections relative to that of random pairs of haplotypes drawn from the entire set of collections. For the analysis among collections within regions,  $\Phi_{SC}$  is the correlation of random haplotypes within collections relative to that of random pairs of haplotypes from the regions. Finally for the within-collection analysis,  $\Phi_{ST}$  is the correlation of random haplotypes within collections relative to that of random pairs of haplotypes drawn from the entire set of collections. The AMOVA analysis allows for only a two-level hierarchy. We will perform two separate analyses, one based on elevation and one based on geographic regions. The significance of the observed variance components and  $\Phi$ -statistics will be tested using a random permutation procedure in AMOVA. The permutation approach to significance testing avoids the parametric assumptions of normality and independence that are not met by molecular distance measures (Excoffier et al. 1992). The number of permutations will be set at 1000 for each analysis.  $\Phi_{ST}$  between pairs of populations, a modified coancestry coefficient, will also be calculated as a genetic distance and examined with multidimensional scaling analysis (MDS; Lessa 1990).

### *Experimental Matings*

In addition to collecting allozyme data from field collections, we will analyze experimental matings to verify the genetic basis of isozyme variation for putative allelic polymorphisms that have not been tested in pink salmon.

In the 1994 and 1995 examination of even-year collections, the subcontractor identified numerous isozyme polymorphisms that were previously undescribed (Table 4). The recently tetraploid salmonids often express an abundance of isozymes from the duplicated loci, and new alleles can initially be difficult to score (cf., Marsden et al. 1987). Difficulty can arise in distinguishing among cryptic variation, single-locus variation from isolocus pairs, and phenotypic variation with a non-genetic basis. The genetic basis and state of duplication for these newly-found polymorphisms must be confirmed before they are incorporated into population structure analyses (e.g., see May et al. 1975; Seeb and Seeb 1986).

The best method to confirm the genetic basis of such polymorphisms is through inheritance studies. We produced single-pair-matings from pink salmon originating in Prince William Sound in 1994 and 1995. We froze tissues from all parents used in these matings. We also froze tissues from 100 progeny from each single-pair-mating and will test parents to identify those families expressing polymorphism for the isozymes listed in

Table 4 in both even and odd years. Inheritance will be determined by scoring phenotypes of the progeny and performing a goodness-of-fit test to Mendelian values expected from both duplicated and non-duplicated loci. Scores for polymorphisms with confirmed genetics basis will be incorporated into the data base for further analyses (above). Joint segregation, if observed, will be reported as a courtesy to the scientific community (cf., May et al. 1982).

### 3. Application to Management

Applying these data to the management of Prince William Sound requires the recognition that diversity must be conserved both within and among populations of pink salmon. The most conservative approach would be to base management on each local spawning aggregation, but our ability to manage on such a fine scale is often limited. Therefore a recognition of the patterns of diversity and the relative amount of diversity distributed at various hierarchical levels is often necessary to devise management strategies that can be implemented.

Recently a management framework for the conservation of genetic diversity was developed by the Washington State Department of Fish and Wildlife (Busack and Marshall 1995). Within this framework, individual stocks are categorized into larger groupings of genetic diversity--*genetic diversity units (GDUs)* and *major ancestral lineages (MALs)* in a hierarchical fashion. This framework is based not only on genetic data, but also geographic and life history information. We propose to follow this framework in our analysis of pink salmon from Prince William Sound. Busack and Marshall (1995) define their lowest grouping as:

A **stock** is a group of interbreeding individuals that is genetically distinct and substantially reproductively isolated from other such groups

Genetic distinctness among stocks can be determined by significant statistical differences in allele frequencies (log-likelihood tests) and, in extreme cases, distinct life history characteristics (e.g. timing of return, upstream vs. tidal spawning location) or geographic separation (Shaklee et al. 1995).

The next hierarchical level is:

A **genetic diversity unit (GDU)** is a group of genetically similar stocks that is genetically distinct from other such groups. The stocks typically exhibit similar life histories and occupy ecologically, geographically, and geologically similar habitats. A GDU may consist of single stock.

This grouping is a relative term; stocks within a GDU are more similar to each other than to stocks in other GDUs. Gene flow among stocks within GDUs may still occur at varying levels. Identification of MALs will be based on results of hierarchical log-likelihood tests, gene diversity analyses, and MDS plots.

The third and highest grouping defined by Busack and Marshall (1995) is:

A **major ancestral lineage** (*MAL*) is a group of one or more genetic diversity units whose shared genetic characteristics suggest a distant common ancestry, and substantial reproductive isolation from other MALs. Some of these groups are likely the result of colonization and diversification preceding the last period of glaciation.

Identification of MALs will also be based on results of hierarchical log-likelihood tests, gene diversity analyses, and MDS plots. Gene flow among MALs would be extremely low or nonexistent.

This approach has been applied to pink salmon from the State of Washington (Shaklee et al. 1995). They identified two major ancestral lineages (MALs) representing even-year and odd-year brood lines. The odd-year consisted of eight genetic diversity units (GDUs) including Nooksack, North Puget Sound, Puyallup, Nisqually, Hood Canal, Upper Dungeness (Summer), Lower Dungeness (Fall), and Hood Canal Hatchery. In contrast, the even-year MAL consisted of a single GDU, Snohomish, even-year.

We believe a similar approach will provide an appropriate framework for the conservation of genetic diversity of pink salmon within Prince William Sound. To the extent possible, ADFG advocates management strategies and genetics policies based upon the "*stock*".



Table 1. Tributaries and hatcheries in Prince William Sound targeted for sampling of odd-year class. Samples were collected opportunistically from 16 spawning aggregates in 1991 and as part of 95196 in 1995. The early, late, upstream, and intertidal aggregations to be sampled in 1997 will be chosen from those listed and will depend on abundance of spawning adults. Physiogeographic characteristics and approximate sampling dates for collecting early- and late-runs are included. Map #'s correspond to numbered locations on Figure 1. Tectonic change is the vertical shift (in meters) resulting from the 1964 earthquake (derived from Plafker and Mayo 1965; isobase map).

Location		Physiographic characteristics		Year	
Map #	Name	Tidal/Upstream	Tectonic change	1991	1995
1	Rocky	Both	+2.4 to + 3.0		8/23
2	Wilby	Tidal	+3.0	8/30	
3	Hayden	Tidal	+3.0	8/18	
4	AFK	Hatchery	+2.4	9/02	
5	Erb	Both	+0.6	8/04*	7/24
				9/05*	8/24
6	Mink	Both	-0.6	7/28*	7/25
					8/25
7	Swanson	Tidal	-1.2 to -1.8	8/06	7/26
					8/26
8	Cannery	Hatchery	0.0	9/12	
9	Long	Tidal	0.0		8/15
10	Solomon G.	Hatchery	0.0	8/08	
				8/20	
11	Duck	Tidal	+0.6 to +1.2	8/20	

Location		Physiographic characteristics		Year	
Map #	Name	Tidal/Upstream	Tectonic change	1991	1995
12	Lagoon	Both	+2	8/02*	7/27 8/27
13	Olsen	Both	+0.6 to +1.2	7/21*	7/28 8/28
14	Koppen	Both	+1.2 to +1.8	9/06* 8/03**	7/29 8/29
15	Humpback	Tidal	+1.8 to +2.4	7/25 8/31	
16	Hartney	Tidal	+1.2 to +1.8	7/31	
17	Constantine	Both	+1.8	8/24*	8/01 9/01

\* Tidal samples only

\*\* Upstream samples only.

Table 2. Enzymes, loci, and their primary tissue-buffer combinations proposed to screen for allozyme variation. Enzyme nomenclature follows Shaklee et al. (1990), and locus abbreviations are given. Buffer abbreviations are as described in the text. These are the same loci and tissue-buffer combinations used in the even-year analysis so the data will be compatible.

Enzyme	Enzyme Number	Locus	Tissue	Buffer <sup>1</sup>
Aspartate aminotransferase	2.6.1.1	<i>sAAT-1,2*</i>	Heart	ACEN 6.8
		<i>sAAT-3*</i>	Eye	TG
		<i>sAAT-4*</i>	Liver	TG
		<i>mAAT-1*</i>	Heart	ACEN 6.8
		<i>mAAT-2*</i>	Muscle	ACE 6.5
Adenosine deaminase	3.5.4.4	<i>ADA-1*</i>	Muscle	AC 6.1
		<i>ADA-2*</i>	Muscle	AC 6.1
Aconitate hydratase	4.2.1.3	<i>mAH-1*</i>	Heart	ACEN 6.8
		<i>mAH-2*</i>	Heart	ACEN 6.8
		<i>mAH-3*</i>	Muscle	ACE 6.8
		<i>mAH-4*</i>	Muscle	ACE 6.8
		<i>sAH*</i>	Liver	ACEN 6.8
Adenylate kinase	2.7.4.3	<i>AK*</i>	Muscle	TG
Alanine aminotransferase	2.6.1.2	<i>ALAT*</i>	Muscle	TG
Creatine kinase	2.7.3.2	<i>CK-A1*</i>	Muscle	TG
		<i>CK-A2*</i>	Muscle	TG
		<i>CK-B*</i>	Eye	TG
		<i>CK-C1*</i>	Eye	TG
		<i>CK-C2*</i>	Eye	TG
Esterase-D	3.1.1.-	<i>ESTD*</i>	Muscle	ACE 6.5
Formaldehyde dehydrogenase	1.2.1.1	<i>FDHG*</i>	Heart	ACEN 6.8
Fumarate hydratase	4.2.1.2	<i>FH*</i>	Muscle	ACE 6.8

Table 2. Continue.

Enzyme	Enzyme Number	Locus	Tissue	Buffer <sup>1</sup>
$\beta$ -N-Acetylgalactosaminidase	3.2.1.53	<i><math>\beta</math>GALA*</i>	Muscle	TG
Glyceraldehyde-3-phosphate dehydrogenase	1.2.1.12	<i>GAPDH-1*</i>	Muscle	AC 6.1
		<i>GAPDH-2*</i>	Heart	ACEN 6.8
Guanine deaminase	3.5.4.3	<i>GDA*</i>	Liver	TG
B-N-Acetyl- $\beta$ -hexosaminidase	3.2.1.53	<i><math>\beta</math>HA*</i>	Liver	ACE 6.8
Glycerol-3-phosphate dehydrogenase	1.1.1.8	<i>G3PDH-1*</i>	Muscle	TG
		<i>G3PDH-2*</i>	Heart	ACEN 6.8
		<i>G3PDH-3*</i>	Heart	ACEN 6.8
Glucose-6-phosphate isomerase	5.3.19	<i>GPI-B1,2*</i>	Muscle	TG
		<i>GPI-A*</i>	Muscle	TG
Glutathione reductase	1.6.4.2	<i>GR*</i>	Heart	TC4
Isocitrate dehydrogenase (NADP+)	1.1.1.42	<i>mIDHP-1*</i>	Muscle	ACE 6.5
		<i>mIDHP-2*</i>	Heart	ACEN 6.8
		<i>sIDHP-2*</i>	Liver	ACE 6.8
L-Lactate dehydrogenase	1.1.1.27	<i>LDH-A1*</i>	Muscle	TG
		<i>LDH-A2*</i>	Muscle	TG
		<i>LDH-B1*</i>	Heart	TG
		<i>LDH-B2*</i>	Heart	TG
		<i>LDH-C*</i>	Eye	TG
Malate dehydrogenase	1.1.1.37	<i>sMDH-A1,2*</i>	Heart	ACEN 6.5
		<i>sMDH-B1,2*</i>	Heart	ACEN 6.5
		<i>mMDH-1*</i>	Heart	ACEN 6.5

Table 2. Continue.

Enzyme	Enzyme Number	Locus	Tissue	Buffer <sup>1</sup>
		<i>mMDH-2*</i>	Heart	ACEN 6.5
Malic enzyme (NADP+)	1.1.1.40	<i>mMEP-1*</i>	Muscle	ACE 6.8
		<i>mMEP-2*</i>	Muscle	ACE 6.8
Mannose-6-phosphate isomerase	5.3.1.8	<i>MPI*</i>	Heart	TG
Nucleoside-triphosphate pyrophosphatase	3.6.1.19	<i>NTP*</i>	Muscle	ACE 6.5
Cytosol non-specific Dipeptidase	3.4.13.18	<i>PEPA*</i>	Muscle	TG
Tripeptide aminopeptidase	3.4.11.4	<i>PEPB-1*</i>	Heart	TG
X-pro-dipeptidase	3.4.13.9	<i>PEPD-2*</i>	Heart	ACEN 6.5
Peptidase-LT	3.4.-.-	<i>PEPLT*</i>	Muscle	TG
Phosphogluconate dehydrogenase	1.1.1.44	<i>PGDH*</i>	Muscle	ACE 6.5
Phosphoglycerate kinase	2.7.2.3	<i>PGK-1*</i>	Muscle	ACE 6.8
		<i>PGK-2*</i>	Muscle	ACE 6.8
Phosphoglucomutase	5.4.2.2	<i>PGM-2*</i>	Heart	TG
Superoxide dismutase	1.15.1.1	<i>sSOD-1*</i>	Heart	ACEN 6.8
		<i>sSOD-2*</i>	Heart	ACEN 6.8
		<i>mSOD*</i>	Heart	ACEN 6.8
Triose-phosphate isomerase	5.3.1.1	<i>TPI-1*</i>	Muscle	TG
		<i>TPI-2*</i>	Muscle	TG
		<i>TPI-3*</i>	Muscle	TG
		<i>TPI-4*</i>	Muscle	TG

<sup>1</sup>Buffers: AC: amine-citric acid buffer, pH 6.8 (Clayton and Tretiak 1972) modified with EDTA (E), NAD (N), or both (Harris and Hopkinson 1976); TBCL: Tris-citric acid gel, pH 8.7 and lithium hydroxide-boric acid electrode buffer, pH 8.0 (Ridgway et al. 1970); TC4: Tris-citric

Table 2. Continue.

acid buffer pH 5.8 (Schaal and Anderson 1974); TG: Tris-glycine buffer, pH 8.5 (Holmes and Masters 1970).

Table 3. Restriction enzymes that were used to screen for RFLP markers in mtDNA during Trustee Council Project 94320D and 95320D. Eighty each of even- and odd-year-class pink salmon from Prince William Sound were initially analyzed. Asterisk indicates enzymes that revealed polymorphism, and these six will be assayed in 40 individuals each from 1997 odd-year class collections for Trustee Council Project 98191.

Restriction Enzyme Screen		Recognition Site
Apa I	*	GGGCC'C
Ase I		AT'TAAT
Ava II		C'YCGRG
Bgl I		GGCNNNN'NGGC
Bgl II		A'GATCT
BstU I *		CG'CG
EcoR V	*	GAT'ATC
Hha I		GCG'C
Hinf I	*	G'ANTC
Mse I		T'TAA
Msp I		C'CGG
Nci I		CC'SGG
RsaI	*	GT'AC
Sac I		GAGCT'C
Sac II		CCGC'GG
Sau96 I		G'GNCC
Sca I		AGT'ACT
Taq I		T'CGA
Xba I	*	T'CTAGA
Xho I		C'TCGAG

Table 4. Putative alleles that will be progeny tested in 1995-1998. Tissue-buffer combinations are those identified by Washington Department of Fish and Wildlife that optimally resolve phenotypes. Alleles expressed as relative mobility to common allele. Buffers: LIOH-R (Ridgway et al. 1970; "UC Davis recipe"); TRIS-MAL7.4 (Shaw and Prasad 1970); TRIS-GLY (Holmes and Masters 1970); TC-4 (Schaal and Anderson 1970, buffer "a"); CAM(E)(N)6.1 and 6.3 (Clayton and Tretiak 1972, (E) = with EDTA, (N) = with NADP). Alleles in **BOLD** are alleles found in our 1994 analysis of even year pink salmon in Prince William Sound that were previously undescribed in pink salmon. Only those previously undescribed alleles associated with loci that have not been subjected to inheritance studies are included.

Locus	Alleles									Tissues	Buffers
	1	2	3	4	5	6	7	8	9		
<i>sAAT-3</i>	100*	91*	<b>79*</b>							E	LIOH-R
<i>AK</i>	-100*	<b>-145*</b>								M	TRIS-GLY
<i>FH</i>	100*	136*								M	TC-4
<i>bGALA</i>	100*	111*	91*	105*						M	TRIS-GLY
<i>GDA</i>	100*	108*	113*	113*	118*	115*	123*	82*	110*	L,M	TRIS-GLY
	100*	130*	155*	100*	189*	167*	222*	93*	106*	L,M	CAM(E)6.8
<i>bGLUA</i>	100*	<b>200*</b>								L	CAMEN6.8

Table 4. Continued

Locus	Alleles									Tissues	Buffers
	1	2	3	4	5	6	7	8	9		
<i>GAPDH-2</i>	100*	127*	87*							M	CAM6.1
<i>G3PDH-2</i>	100*	120*	90*							H	CAMEN6.8
<i>G3PDH-3</i>	100*	90*								H	CAMEN6.8
<i>IDDH-1</i>	100*	134*								L	LION-R
<i>LGL</i>	100*	80*								M,H	TRIS-GLY
<i>aMAN</i>	100*	85*								H	TRIS-GLY
<i>mMDH-2,3</i>	100*	228*								H,M	CAME(N)6.8
<i>NTP</i>	100*	53*	130*							M,L	CAME6.8
<i>mSOD</i>	100*	145*	14*	185*	118*	69*				H	TC-4
<i>sSOD-2</i>	100*	122*								H	CAM6.1



Figure 1. Locations for sampling odd-year pink salmon in Prince William Sound and isobases indicating vertical shift (in feet) resulting from the 1964 earthquake. Numbers on map correspond to Map # on Table 1.

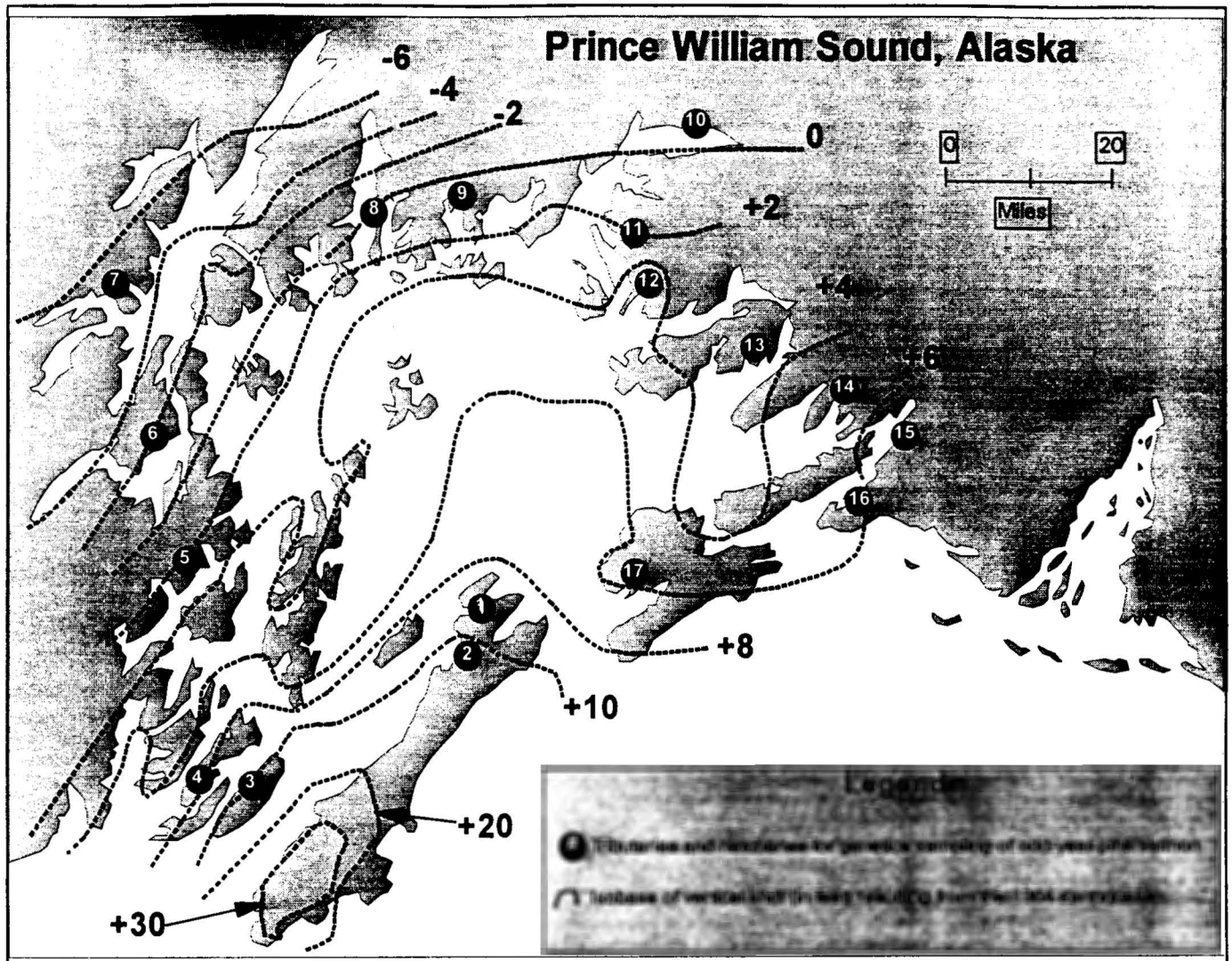
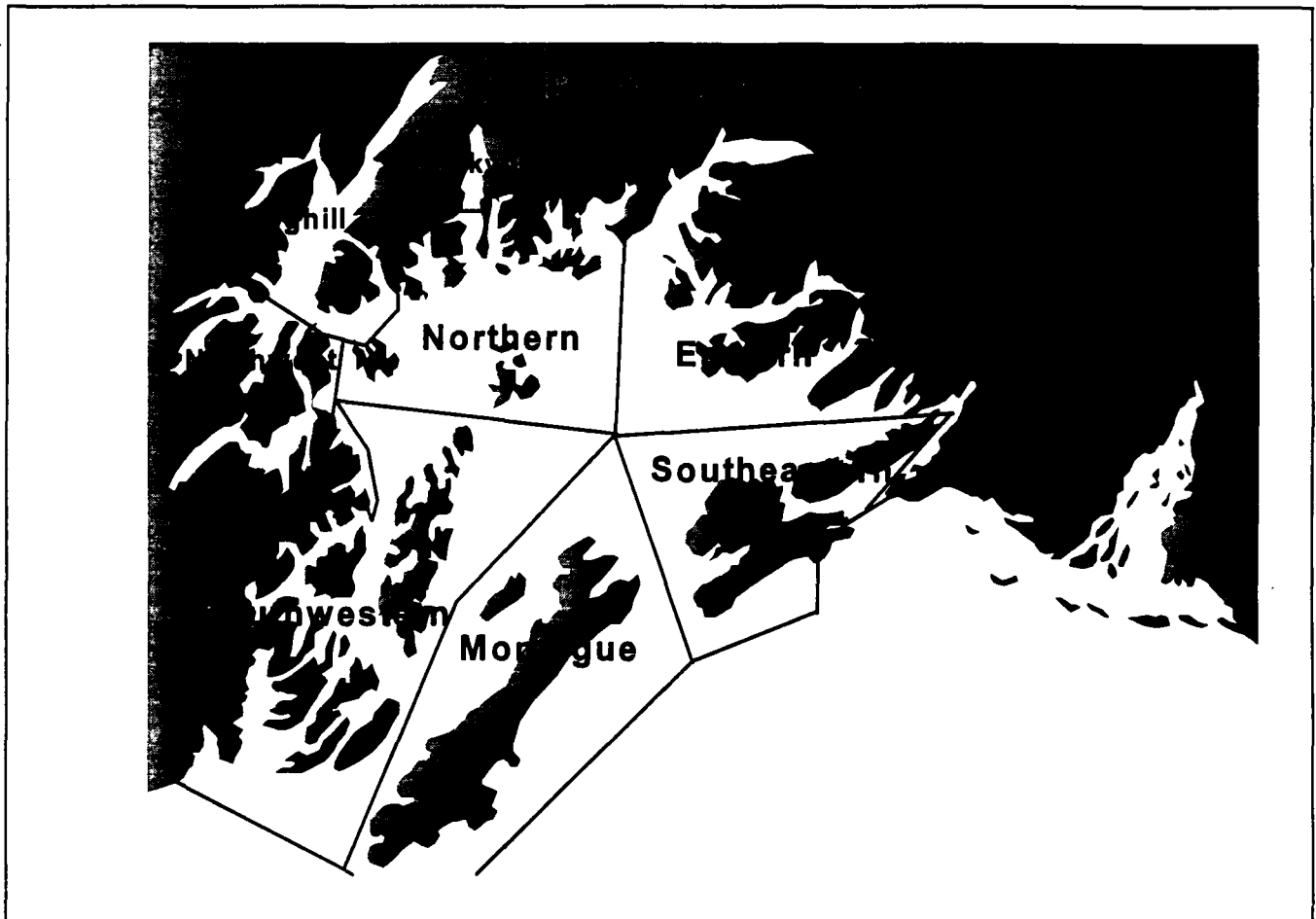


Figure 2. Management districts in Prince William Sound, AK. In this study, four of the smaller districts were incorporated into the five major districts (Southeast, East, North, Southwest, Montague). Three of these (Unakwik, Coghill, and Eshamy) were originally delimited for sockeye salmon management and will be combined into the North (Unakwik and Coghill) and Southwest (Eshamy) districts for this study. The Northwest district, a small region originally delineated for chum salmon and pink salmon management, will be combined into the North district for this study because of its small size, geographic location, and run timing similarities of pink salmon inhabiting these two districts.



### **C. Cooperating Agencies, Contracts and Other Agency Assistance**

Because of the 1994 State of Alaska hiring freeze, Alaska Department of Fish and Game Genetics Laboratory subcontracted the allozyme portion of Project 94320D to Washington Department of Fish and Wildlife as the best-qualified bidder identified through the state procurement process. The soundness of this decision was confirmed through the peer review of Project 95320D by independent consultants to the Chief Scientist of the Trustee Council.

However, the cost of the subcontract to WDFW in 1994 exceeded the total amount awarded by the Trustees to the ADFG Genetics Lab. This cost increase was due to many factors including: (1) elevated costs of performing the work outside of ADFG, (2) accommodation of Project 94320D to peer-review recommendations for increased analysis of stocks in southwestern PWS to test outbreeding-depression hypothesis (to explain embryo mortalities observed in results of Trustee Council Project 94191), and (3) a decision to add additional loci to the locus screen made by the principal investigator (JES) as a result of negotiations with the subcontractor. ADFG handled the resulting budget problem internally in FY 94 by appropriately supplementing the subcontract with funds from Trustee Council Project 95191 and by postponing some of the ADFG mtDNA analyses until FY 95.

These contractual shortfalls were ameliorated in the budget for FY 95. That budget included a subcontract for continued work by WDFW for the analyses of 2000 samples of odd-year origin. The provision for this contract-extension was included in the terms of the 1994 award to WDFW. We have contracted the allozyme portion of project 96196 to Washington Department of Fish and Wildlife to analyze the 1995 samples.

We intend to perform the allozyme analysis of the 1996 and 1997 samples in-house. The 1996 samples will be done in FY 97 under project 97196. Therefore, budgets for FY 97 and beyond reflect costs for analysis of allozyme samples in Anchorage at the ADFG facility.

## **SCHEDULE**

### **A. Measurable Project Tasks for FY 98 (October 1, 1997 - September 30, 1998)**

Oct. 1997:	Finish allozyme lab analysis of 1996 collections
Oct. - Dec 1997:	Finish mtDNA analysis of 1996 collections
Oct. 1997 - May 1998:	Allozyme lab analyze 1997 collections
Nov. 1997 - Jan 1998:	Statistically analyze 1996 collections
Jan 22-25 1998	Attend the Annual Restoration Workshop
Jan - June 1998:	mtDNA analysis of 1997 collections
Feb.- April 1998:	Write-up 1996 results
April 1998:	Final report of FY 97 results - 96 collections, 95 matings
June - Aug. 1998:	Allozyme lab analyze experimental matings
July - Sept. 1998:	Statistically analyze 1997 collections and 1996 matings

## **B. Project Milestones and Endpoints**

October 31, 1997: Complete allozyme lab analysis of 1996 collections  
December 30, 1997: Complete mtDNA lab screen of 1996 collections  
April 15, 1998: Complete evaluation of population structure for 1994-1996 collections  
Sept 30, 1998: Complete screen of samples collected during 1997  
April 30, 1999: Evaluation of population structure of Prince William Sound and other related spawning aggregates collected through 1997  
December 30, 1999: Complete screen of samples collected 1998  
April 15, 2000: Complete evaluation of stability of population structure across years

## **C. Completion Date**

All project objectives will be met in FY 99

## **PUBLICATIONS AND REPORTS**

April 15, 1998: Annual report for FY 97  
September 30, 1999: Final project report in the form of manuscript submitted to journal

Manuscripts funded by this project:

Seeb, J. E. C. Habicht, J. B. Olsen, and L. W. Seeb. An overview of gene detection methods used to study population variation in salmonids. Assessment and Status of Pacific Rim Salmonid Stocks. North Pacific Anadromous Fish Commission, Vancouver B.C. *Accepted and in press.*

Seeb, J. E., C. Habicht, J. B. Shaklee, and L. W. Seeb. Allozymes and mtDNA describe population structure of even-year pink salmon (*Oncorhynchus gorbuscha*) affected by the Exxon Valdez oil spill in Prince William Sound. *In ADFG internal review.*

Habicht, C., S. Sharr, and J. E. Seeb. Coded wire tag placement affects homing ability of pink salmon. Transactions of the American Fisheries Society. *Accepted and in press.*

Fetzner J. W., L. W. Seeb, and J. E. Seeb. Discrimination of even-and odd-year pink salmon (*Oncorhynchus gorbuscha*) populations from Alaska using restriction site variation from the mitochondrial ND5/6 genes. *Submitted to Molecular Ecology.*

Olsen, J. B., J. K. Wenburg, and P. Bentzen. 1996. Semiautomated multilocus genotyping of Pacific salmon (*Oncorhynchus* spp.) using microsatellites. Molecular Marine Biology and Biotechnology. 5:259-272.

Olsen, J. B., J. E. Seeb, L. W. Seeb. Genetic variation at microsatellite loci in North American

odd-year pink salmon (*Oncorhynchus gorbuscha*). Submitted to *Transactions of the American Fisheries Society*.

## **PROFESSIONAL CONFERENCES**

AFS National Meeting - Santa Monica, CA - August 1998 - present paper on results through FY 97 from this project.

AFS Alaska chapter - Juneau, AK - November 1997 - present paper on results through FY 96 from this project.

## **NORMAL AGENCY MANAGEMENT**

The need for characterization the genetic structure of pink salmon within the Sound has increased as a direct result of the EVOS. Western PWS stocks were directly impacted by the oil spill as discussed in Craig et al. (1996) and Miller et al. (1994). In addition, eastern PWS stocks were depleted following the spill as a result of a shift in harvest pressure from western to eastern stocks in 1989. In order to restore these damaged stocks, supplementation projects often are proposed to the Exxon Valdez Trustee Council. Understanding of stock structure is critical to assess potential genetic impacts such projects would have on wild pink salmon (Trustee Council Projects R105, 95320 A-P, 95093). Additionally, managing the harvest of pink salmon in areas where wild populations were damaged by the spill would be benefited by a better understanding of the stock structure because this understanding will provide managers with the appropriate scale for fisheries management.

Characterization of the genetic structure of pink salmon within PWS was not high enough on the Department's priority to have occurred before EVOS. However, once the data has been collected it will be useful to the Department for future management of pink salmon within PWS and the database will be maintained and updated by the Department after the project funding ends.

The Department is demonstrating its commitment to this project by fully funding the project leaders: Christopher Habicht, James Seeb, and Lisa Seeb.

## **COORDINATION AND INTEGRATION OF RESTORATION EFFORT**

Previous assessments of egg and fry survival in oiled and unoiled streams demonstrated detrimental effects of EVOS on pink salmon (Natural Resources Damage Assessment Fish/Shellfish Study # 2 *Injury to Salmon Eggs and Preemergent Fry* and EVOS Trustee Council Projects R60C, 93003, and 94191 *Oil Related Egg and Alevin Mortalities*). The heritable, genetic nature of the damage was revealed in matings performed as a part of Project 93003. In response to those findings, coded-wire tag recoveries from pink salmon in PWS (e.g., Natural Resources

Damage Assessment Fish/Shellfish Study # 3 and Projects R60A and 93067) were used to reduce the fishing effort on wild pink salmon "populations" through fisheries management. Yet the actual genetic structure of pink salmon populations in PWS remains unknown.

Therefore, Trustee Council Project 98196 was designed to provide a genetic basis for the hatchery/wild-stock components of Project 98320 *Prince William Sound Ecosystem Investigation* and to provide the information essential for population-specific management through such projects as 94184 Coded-Wire-Tag *Recoveries from Pink Salmon in Prince William Sound Fisheries* and others that may be proposed as a consequence of 98320.

## **EXPLANATION OF CHANGES IN CONTINUING PROJECTS**

This year (97196) is the last year that we collect field samples. This change is reflected in the reduced budget request for 98196.

## **PROPOSED PRINCIPAL INVESTIGATOR**

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## **PRINCIPAL INVESTIGATOR**

**Christopher Habicht, Fisheries Biologist II**  
Commercial Fisheries Management and Development  
Alaska Department of Fish and Game  
Anchorage, Alaska 99518 (907) 267-2385

PROJECT RESPONSIBILITIES: Laboratory analysis, data analysis, reporting.

## **EDUCATION:**

B.S., 1986, Fisheries Science, Cornell University, Ithaca NY  
M.S., 1994, Zoology, Southern Illinois University, Carbondale IL

## **PROFESSIONAL EXPERIENCE:**

1992- Fisheries Biologist, C.F.M.D. Division, ADFG

Supervising laboratory analysis of genetic markers for EVOS Trustee Council study 93012 (Genetic Stock Identification of Kenai River Sockeye Salmon). Conducting laboratory evaluations of genetically altered salmonids. Analyzing straying data from pink salmon and chinook salmon tag recoveries.

- 1989-1992 Graduate Assistant, Southern Illinois University  
Conducted allozyme species identification, developed *in vivo* ova storage techniques, and optimized triploid induction and gynogenesis protocols for moronids.
- 1986-1989 Research Associate, Ohio State University  
Provided field and laboratory support for aquatic ecology studies on bioenergetics of essocids.

#### PUBLICATIONS AND PRESENTATIONS:

- Habicht, C. 1993. Electrophoretic Identification of *Morone* species, and *In Vivo* ova storage, induced gynogenesis, and induced triploidy in white bass (*M. chrysops*). Masters Thesis, Southern Illinois University, Carbondale IL.
- Seeb, L. W., J. E. Seeb, C. Habicht. 1993. Population genetic analyses facilitate restoration of sockeye salmon stocks damaged by the *Exxon Valdez* oil spill. Presented at National Chapter American Fisheries Society, Portland, OR.
- Habicht, C. 1994. Gene conservation of triploids in the management of salmonids. Presented at North American Fish and Wildlife Conference, Anchorage, AK.
- Habicht, C., J. E. Seeb, R. B. Gates, I. R. Brock, and C. A. Olito. 1994. Triploid salmon outperform diploid and triploid hybrids between coho salmon and chinook salmon during their first year. *Can. J. Fish. Aquat. Sci.* 51(Suppl. 1):31-37.
- Seeb, L. W. C. Habicht W. D. Templin, K. E. Tarbox R. Z. Davis, L. K. Brannian, J. E. Seeb. *accepted*. Genetic diversity of sockeye salmon (*Oncorhynchus nerka*) of Cook Inlet, Alaska, and its application to restoration of populations affected by the *Exxon Valdez* oil spill. *Canadian Journal of Fisheries and Aquatic Sciences*. 00:000-000.
- Seeb, J. E. C. Habicht, J. B. Olsen, and L. W. Seeb. *in press*. An overview of gene detection methods used to study population variation in salmonids. *Assessment and Status of Pacific Rim Salmonid Stocks*. North Pacific Fish Commission, Vancouver B.C.
- Habicht, C., S. Sharr, and J. E. Seeb. *accepted*. Coded wire tag placement affects homing ability of pink salmon. *Transactions of the American Fisheries Society*. 00:000-000.

## OTHER KEY PERSONNEL

### A. James E. Seeb, Principal Geneticist

Commercial Fisheries Management and Development  
Alaska Department of Fish and Game  
Anchorage, Alaska 99518 (907) 267-2385

PROJECT RESPONSIBILITIES: Design, analysis, reporting

### EDUCATION:

B.S., Biology, 1974, University of Puget Sound  
M.S., Fisheries, 1982, University of Washington  
Ph.D., Fisheries, 1987, University of Washington

### PROFESSIONAL EXPERIENCE:

1990- Principal Geneticist, CFMD Division, ADFG  
1991- Affiliate Associate Professor, U. of Alaska, Fairbanks  
1988-1990 Assistant Professor, Southern Illinois University  
1987-1988 Research Assistant Professor, University of Idaho  
1982-1986 Graduate Research Assistant, University of Washington  
1980-1982 Fish Biologist, Pacific Fisheries Research, Olympia, WA  
1978-1980 Fish Biologist, Washington Department of Fisheries

### SELECTED PUBLICATIONS:

Seeb, J. E., L. W. Seeb, and F. M. Utter. 1986. Use of genetic marks to assess stock dynamics and management programs for chum salmon. *Trans. Amer. Fish. Soc.* 115:448-454.

Seeb, J. E., and L. W. Seeb. 1986. Gene mapping of isozyme loci in chum salmon (*Oncorhynchus keta*). *J. Hered.* 77:399-402.

Seeb, J. E., L. W. Seeb, D. W. Oates, and F. M. Utter. 1987. Genetic variation and postglacial dispersal of populations of northern pike (*Esox lucius*) in North America. *Can. J. Fish. Aquat. Sci.* 44:556-561.

Utter, F. M., and J. E. Seeb. 1990. Genetic marking of fishes: overview focusing on protein variation. *Am. Fish. Soc. Sym.* 7:426-438.

Seeb, J. E., G. H. Kruse, L. W. Seeb, and R. J. Weck. 1990. Genetic structure of red king crab populations in Alaska facilitates enforcement of fishing regulations. *Proceedings of the International Symposium on King and Tanner Crabs. Alaska Sea Grant, Fairbanks, AK.* pp 491-502.



- Seeb, J. E., and G. D. Miller. 1990. The integration of allozyme analyses and genomic manipulations for fish culture and management. *In*: D.H. Whitmore, Editor. Electrophoretic and Isoelectric Focusing Techniques in Fisheries Management. CRC Press, Boca Raton, pp 266-279.
- Seeb, J. E., C. Habicht, J. Fetzner, W. Templin, L. W. Seeb. 1996. Allozymes and mtDNA describe population structure of even-year pink salmon (*Oncorhynchus gorbuscha*) affected by the *Exxon Valdez* oil spill in Prince William Sound. Exxon Valdez Oil Spill Restoration Project Annual Report Restoration Projects 94320D and 95320D, Alaska Dept. of Fish and Game, Anchorage, Alaska.
- Seeb, J. E., G. H. Thorgaard, and T. Tynan. 1993. Triploid hybrids between chum salmon female x chinook salmon male have increased seawater tolerance. *Aquaculture* 117:37-45.
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- Crane, P. A., L. W. Seeb, and J. E. Seeb. 1994. Genetic relationships among *Salvelinus* species inferred from allozyme data. *Can. J. Fish. Aquat. Sci.* 51(Suppl. 1):182-197.
- Seeb, L. W. C. Habicht W. D. Templin, K. E. Tarbox R. Z. Davis, L. K. Brannian, J. E. Seeb. *accepted*. Genetic diversity of sockeye salmon (*Oncorhynchus nerka*) of Cook Inlet, Alaska, and its application to restoration of populations affected by the *Exxon Valdez* oil spill. *Canadian Journal of Fisheries and Aquatic Sciences*. 00:000-000.
- Seeb, J. E. C. Habicht, J. B. Olsen, and L. W. Seeb. *in press*. An overview of gene detection methods used to study population variation in salmonids. *Assessment and Status of Pacific Rim Salmonid Stocks*. North Pacific Fish Commission, Vancouver B.C.
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- Fetzner J. W., L. W. Seeb, and J. E. Seeb. *submitted to Molecular Ecology*. Discrimination of even-and odd-year pink salmon (*Oncorhynchus gorbuscha*) populations from Alaska using restriction site variation from the mitochondrial ND5/6 genes.

**B. Lisa W. Seeb (L. Wishard), Statewide Geneticist**  
 Division of Commercial Fisheries Management and Development  
 Alaska Dept. of Fish and Game  
 Anchorage, Alaska 99518 (907) 267-2249

PROJECT RESPONSIBILITIES: Biometrics, analysis, reporting

## EDUCATION:

A.B. Zoology, 1973, University of California, Berkeley

M.A. Zoology, 1977, University of Montana

Ph.D. Fisheries, 1986, University of Washington

## PROFESSIONAL EXPERIENCE:

1991- Statewide Geneticist, ADFG, Anchorage

1991- Affiliate Associate Professor, U. of Alaska, Fairbanks

1988-1990 Assistant Professor, Southern Illinois University

1984-1988 Research Assist. Prof., University of Idaho

1978-1981 Fish Geneticist, Pacific Fish. Research, Olympia WA

1977-1979 Geneticist, National Marine Fisheries Service, Seattle

## SELECTED PUBLICATIONS:

Wishard, L. N., J. E. Seeb, F. M. Utter, and D. Stefan. 1984. A genetic investigation of suspected redband trout populations. *Copeia* 1984(1):120-132.

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Seeb, L. W., J. E. Seeb, R. L. Allen and W. K. Hershberger. 1990. Evaluation of adult returns of genetically marked chum salmon, with suggested future applications. *American Fisheries Society Symposium* 7:418-425

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Seeb, L. W., J. E. Seeb and J. J. Polovina. 1990. Genetic variation in highly exploited spiny lobster *Panulirus marginatus* populations from the Hawaiian Archipelago. *Fishery Bulletin* 88:713-718.

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# 1998 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1997 - September 30, 1998

<b>Budget Category:</b>	Authorized FY 1997	Proposed FY 1998						
	\$121.4							
Personnel	\$8.8	\$92.8						
Travel	\$16.0	\$0.8						
Contractual	\$30.0	\$3.0						
Commodities	\$0.0	\$19.5						
Equipment	\$176.2	\$0.0	LONG RANGE FUNDING REQUIREMENTS					
Subtotal	\$19.3	\$116.1		Estimated FY 1999	Estimated FY 2000	Estimated FY 2001	Estimated FY 2002	
General Administration	\$195.5	\$14.1						
Project Total	\$214.8	\$130.2		\$50.0	\$0.0	\$0.0	\$0.0	
Full-time Equivalents (FTE)		2.3						
Dollar amounts are shown in thousands of dollars.								
Other Resources								
Comments:								

**1998**

Project Number: 98196  
Project Title: Pink Salmon Stock Genetics  
Agency: ADF&G

FORM 3A  
TRUSTEE  
AGENCY  
SUMMARY

**1998 EXXON VALDEZ TRUSTE JUNCIL PROJECT BUDGET**

October 1, 1997 - September 30, 1998

<b>Personnel Costs:</b>		GS/Range/ Step	Months Budgeted	Monthly Costs	Overtime	Proposed FY 1998
Name	Position Description					
	FWT III	11A	11.0	3.32		36.5
	FWT II	9F	11.0	3.40		37.4
	FWT II	9C	6.0	3.15		18.9
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
Subtotal			28.0	9.9	0.0	
<b>Personnel Total</b>						<b>\$92.8</b>
<b>Travel Costs:</b>		Ticket Price	Round Trips	Total Days	Daily Per Diem	Proposed FY 1998
Description						
1 Round trip to scientific meeting		0.8	1			0.8
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
<b>Travel Total</b>						<b>\$0.8</b>

**1998**

Project Number: 98196  
Project Title: Pink Salmon Stock Genetics  
Agency: ADF&G

FORM 3B  
Personnel  
& Travel  
DETAIL

# 1998 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1997 - September 30, 1998

Contractual Costs:		Proposed
Description		FY 1998
Photography		2.0
Publication		1.0
When a non-trustee organization is used, the form 4A is required.		
<b>Contractual Total</b>		<b>\$3.0</b>
Commodities Costs:		Proposed
Description		FY 1998
Biochemicals		15.0
Miscellaneous laboratory supplies		3.5
Office supplies		1.0
<b>Commodities Total</b>		<b>\$19.5</b>

**1998**

Project Number: 98196  
Project Title: Pink Salmon Stock Genetics  
Agency: ADF&G

FORM 3B  
Contractual  
& Commodi  
ties

### COUNCIL PROJECT BUDGET

October 1, 1997 - September 30, 1998

[illegible]

## 1998

Project Number: 98196  
Project Title: Pink Salmon Stock Genetics  
Agency: ADF&G

FORM 3B  
Equipment  
DETAIL

Prepared: 4 of 4

4/15/97



## Youth Area Watch

Project Number: 98210

Research Category: General Restoration

Proposer: Chugach School District

Lead Trustee Agency: ADFG

Cooperating Agency: DNR

Alaska SeaLife Center: Yes

Duration: 3<sup>rd</sup> year, seven year project

Cost FY 98: \$150,200

Cost FY 99: \$175,000

Cost FY 00: \$175,000

Cost FY 01: \$175,000

Cost FY 02: \$175,000

**RECEIVED**  
APR 11 1997

**EXXON VALDEZ OIL SPILL  
TRUSTEE COUNCIL**

Geographic Area: Prince William Sound and Resurrection Bay  
including: Cordova Harbor and Orca Inlet, Port San  
Juan and Evans  
Island, Tatitlek Narrows, Boulder Bay and  
Landlocked Bay.

### ABSTRACT

Youth Area Watch links students in the oil spill impacted area with research and monitoring projects funded through the Trustee Council. The goal is to involve students in the restoration process, and give these individuals the skills to participate in oil spill restoration activities now and in the years to come. Youth conduct research identified by principal investigators who have indicated interest in working with students in oil spill impacted communities. To instill a long-term commitment to the goals set out in the restoration plan adopted by the

Trustee Council, coordination between current projects and the communities and youth populations will ensure successful results. Youth Area Watch serves as a positive example of community investment in the restoration process. Participating communities include: Tatitlek, Chenega Bay, Cordova, Seward, Valdez, and a remote site within the Chugach School District.

## INTRODUCTION

In the year and a half that Youth Area Watch has operated in the communities, coordination between research and restoration projects and the communities affected by the oil spill has increased. Resulting from many factors, community involvement in the restoration process continues to grow and strengthen; Youth Area Watch is an example of this coordinated effort through the connection that students, the communities and researchers maintain. This relationship provides an environment where youth are encouraged to interpret the data collected and apply the information to the ecosystem.

Students from the oil spill impacted communities are screened and selected for participation in Youth Area Watch at the beginning of the school year. Those showing an interest, academic ability and concern for the oil spill effects on local ecosystems are invited to represent their community as a student of the project. Students work with principal investigators of research projects and community facilitators, as well as independently to achieve the set project objectives.

Four core research projects funded by the Trustee Council serve as central link for all Youth Area Watch activities. Initial cooperating projects include pristane mussel analysis (97195), harbor seal management and biological sampling (97244F), oceanographic data collection (97320M and 97320H), fish monitoring (97320E, 97320T and 97320U). These projects continue to work with Youth Area Watch, providing specific research activities for students to conduct and training protocol for those duties. According to protocol, students collect samples and data for the cooperating research and monitoring projects. The samples and data are compiled by the Youth Area Watch project coordinator located in Anchorage and sent on to the principal investigator of the respective projects. Information on the data collected is maintained by the project coordinator for project analysis conducted by the students during group project sessions.

During the second year of Youth Area Watch, individual community projects began working closely with the students. Those students participating in Youth Area Watch identified a local community restoration project which they could work with. In addition, an open invitation for the participation of other general research and restoration project continues.

## **NEED FOR THE PROJECT**

### **A. Statement of Problem**

Youth Area Watch, identified by the Trustee Council as a "general restoration" project, is committed to collecting the requisite samples and data for principal investigators of research projects to make informed decisions concerning the ecology of oil spill impacted areas. Research and restoration project PI's identify needed data collection within the oil spill impacted communities that in many instances can best be facilitated through local involvement of community residents.

Given the finite resources available for project activities, cost containment is necessary. By working with local community youth, information can be collected at a minimal cost. In addition, a greater quantity of data from an increased number of sites throughout the year can be accomplished by Youth Area Watch project activities.

As a part of the Memorandum of Agreement and Consent Decree approved by the U.S. District Court, "meaningful public participation in the injury and assessment and restoration process" is recognized as an important component of the restoration process. While there are a variety of instituted mechanisms for this involvement, Youth Area Watch offers positive examples of meaningful public participation expressed by the oil spill impacted communities through the involvement of community facilitators (Community Involvement \052A) and other community-based projects. The 1997 Science Review also provided anecdotal, yet substantive support for what the project has done for the restoration process and the communities that are impacted.

### **B. Rationale/Link to Restoration**

Youth Area Watch is based on the commitment of research and restoration projects to involvement students in their work. Participating projects are funded by the Trustee Council and have met the guidelines under the settlement. It is through the cooperating projects that Youth Area Watch holds an interest in the immediate restoration activities.

As a long-term goal, project activities are expected to provide the foundation for long-term claim to restoration of the impacted area to pre-spill levels. Involvement of youth in research and monitoring activities is essential to developing local commitment to the restoration plan adopted by the Trustee Council. Already, students have a greater appreciation for their work as it relates to research projects, as cooperating PI's have requested precise and detailed sampling/data collection protocol from the youth. The youth, in turn,



have increased their knowledge and participation level of community projects. As a result, students now hold a stake in the projects they participate in.

### **C. Location**

While Youth Area Watch is administered through the Chugach School District's main office in Anchorage by the project coordinator, project activities currently take place in the five participating communities, a remote site and in the oil spill impacted area. Local communities include Chenega Bay, Cordova, Seward, Tatitlek and Valdez.

The science teacher within each of the five communities oversees the day-to-day activities pertaining to the project. The project coordinator travels to the local communities to facilitate the off-shore research in specific locations of importance to the identified research projects. Local projects activities identified by each site occur at or near the community. In the case of the remote site, the project coordinator and a principal investigator travels to the student to work one-on-one and provide periodic oversight.

## **COMMUNITY INVOLVEMENT AND TRADITIONAL ECOLOGICAL KNOWLEDGE**

One of the main goals of Youth Area Watch is to facilitate community involvement of the restoration process at a primary school age. It is through community interest and participation that the project has had a positive impact on students. As a result, communities continue to request to participate in Youth Area Watch.

Local oil spill impacted communities are involved and participate in Youth Area Watch. The local facilitators of Community Involvement (/052A) continue to work closely with students and the community Youth Area Watch activities to involve youth where appropriate. Local facilitators and parents of participating youth assist with various aspects of project activities such as serving as chaperones, providing tradition ecological knowledge and coordinating opportunities for youth to work with local projects. Through this cooperative effort, information is exchanged between projects and across generations.

As a component of the second year project scope, students at each site were asked to identify a local project that they could work with. Through these local projects, students gain a greater understanding of what the research and restoration process means at the community level, as well as an interest in

meaningful project outcomes.

## **PROJECT DESIGN**

### **A. Objectives**

Selected students from the identified communities participate in research and restoration activities set out by the Sound Ecosystem Assessment (SEA) Program principal investigators, NOAA staff, Chugach Forest Service biologists and other project principal investigators working with Youth Area Watch. As part of an area watch project that works with existing research and restoration projects. Students collect samples and data that is then provided to the respective projects.

Youth Area Watch objectives are:

1. Selecting students for participation in Youth Area Watch.
2. Finalizing memoranda of agreement with cooperating school districts (for participating sites outside of the Chugach School District).
3. Identifying local research and restoration projects that sites will work on.
4. Completion of protocol for students for all cooperating projects.
5. Providing teacher orientation and student training.
6. Conducting fish monitoring activities and collection of samples/data.
7. Conducting oceanographic data collection.
8. Collecting samples for pristane/mussel analysis.
9. Working with hunters to collect harbor seal samples.
10. Identifying and conducting a local research/restoration activity in each community site.
11. Working with principal investigators to gain awareness of the ecosystem and provide information for research and restoration.

### **B. Methods**

The Chugach School District currently works with the Kenai Peninsula Borough School District, Cordova School District and Valdez School District through memoranda of agreement to allow the communities of Chenega Bay, Cordova, Seward, Tatitlek and Valdez to participate. School districts will operate under the existing agreements during the third project year.

The Youth Area Watch project coordinator works with the principal investigators of the cooperating projects to solidify project expectations. Protocol is established for sample/data analysis. In addition, principal investigators commit to working with the students for a period of time during the training and/or data collection stage.

The Chugach School District developed an application and screening tool to select students for participation in the project. Up to 28 students will be selected from the communities to be a part of Youth Area Watch. While the distribution may vary according to the interest and ability of students that apply, it is expected that the distribution will be as follows: four students from Chenega Bay, four students from Cordova, eight students from Seward, four students from Tatitlek, four students from Valdez, one student from Whittier and one remote site student.

Prior to the beginning of school in the fall, participating Youth Area Watch teachers at the local sites will come together for an orientation session facilitated by the project coordinator. It is anticipated that site teachers will again receive protocol training directly from principal investigators. This training will occur at one community site and the training will be videotaped for future referral. Where principal investigators are not able to attend, protocol training will be arranged informally, yet through direct contact with the site teachers.

Youth Area Watch relies on the participation of research projects, sites and program resources to successfully fulfill the project objectives. Throughout the project year, students travel to research vessels, specific project sites near their community and to research labs in the process of project activity completion. In the past year, Youth Area Watch was able to coordinate with projects conducting research cruises, work cooperatively on task completion while sharing the costs of vessel hiring. In one instance, the principal investigator was unable to make it to the vessel and had the students complete project activities in their stead. It is expected that this type of cooperative effort will continue in the present and coming years.

Students will participate in the four core research projects as a group. This will consist of coming together as a group to work on collection protocol, as well as conducting activities for these projects in their community. In addition, students will participate in local projects that pertain to their geographic area; it is during the local project work that students receive a high degree of one-on-one interaction and involvement with principal investigators and their research.

Ongoing Youth Area Watch research and restoration projects include:

1. Pristane/mussel analysis, Project Number 97195. Jeff Short and Pat Harris at the NOAA Auke Bay laboratory study the pristane levels in blue mussels. There are approximately thirty mussel collection sites in Prince William Sound. Students will continue to collect mussels twice a month at sites appropriate for collection according to set protocol. During the fall and

winter months, students are responsible for overall mussel bed seasonal watch. Students will tag, identify mussel bed characteristics and predator/prey activities.

2. Harbor seal management and biological sampling, Project Number 97244F. The project is conducted by Monica Reidel of the Alaska Native Harbor Seal Commission, in cooperation with Kate Wynne of the University of Alaska, Fairbanks. After they have participated in traditional ecological knowledge and protocol training, students will pair up with local technicians/hunters and assist with bio-sampling activities. Students collect different parts of the seal, including the skin, blubber, teeth and stomach. Adherence to sampling protocol is ensured by working directly with the local hunters.
3. Oceanographic data collection. Observational Physical Oceanography in Prince William Sound, Project Number 97320M. The Role of Zooplankton in Prince William Sound Ecosystem. Shari Vaughan is the principal investigator for these projects. Physical oceanography activities will include measuring basic oceanographic features such as temperature, salinity and weather conditions. Research activities include, 1) temperature: reversing thermometer units and a temperature logger will be monitored by students at research sites, 2) temperature and salinity: CTD (conductivity, temperature and depth) instruments are operated by students to download data into a computer database, and 4) weather station: weather station instruments are installed at each site so that students can measure wind speed and direction, are temperature and barometric pressure.

Students will also collect zooplankton samples as part of on-going SEA biological oceanographic research. Zooplankton collections occur at selected sites to increase the sample range of 97320H. Collection nets are made available from the Zooplankton project.

4. Fish monitoring: Project Number 97320E, Juvenile Salmon Growth and Mortality; Project Number 97320T, Juvenile Herring Growth and Habitats; and Project Number 97320U, Pollock and Herring Energetics. Evelyn Brown works with the Juvenile Herring project and coordinates with the other fish monitoring projects. She provides the protocol that students follow for these project activities.

In addition to the four core projects that Youth Area Watch students participates in, each site is selecting a restoration project to work on in their local community. This restoration activity is something that the students select and not necessarily a project that is currently funded by the Trustee Council. However, local projects are closely linked to existing restoration activities.

Community restoration activities include octopus tagging and general monitoring in Cordova, Anderson Creek enhancement in Chenega Bay and

working with the clam project in Tatitlek; Seward students work with the Institute of Marine Science studying energetics; and the Whittier student will be conducting stream enhancement at Portage Creek. Valdez has not selected a local activity to date, yet a project will be identified before FY 98.

During FY 97, coordination between Youth Area Watch and participating research projects increased significantly. Where possible, research vessel costs were shared to maximize resources for project activities. In the case of the pristine/mussel project, Youth Area Watch paid for the biologist's charter to sites for mussel collection to allow students to participate in the process. Cost sharing also continues when hiring research vessels.

### *Objectives and Activities*

Objective 1: Students are selected to participate in Youth Area Watch.

Activity 1: The project coordinator distributes the student application to project sites. All village council/tribal offices (Chenega Bay, Seward, Tatitlek, Valdez) will receive application forms, as well as the Valdez, Cordova and Kenai Peninsula Borough School Districts for their respective community sites.

Activity 2: The project coordinator will convene a committee to review student applications for Youth Area Watch participation. The committee will be comprised of Chugach School District staff and may be assisted by participating school district staff and community facilitators (/052).

Activity 3: The review committee will select students according to their application reflecting science interests, academic achievement, maturity and site teacher recommendation.

Objective 2: The project coordinator will update memoranda of agreement with the Valdez School District, Cordova School District, and Kenai Peninsula Borough School District for participation in Youth Area Watch.

Activity 1: The project coordinator will contact each school district to evaluate the current agreement, make any necessary changes.

Activity 2: The site teacher will be identified by each school district for the participating communities.

Objective 3: - The project coordinator will identify all research and data collection activities to be conducted by students at all sites participation in Youth Area Watch.

Activity 1: The project coordinator will meet with the principal investigators or delegate project research personnel either by phone or in person by the end of the first fiscal year project month to set student activity parameters.

Activity 2: Activity protocol will be forwarded by the principal investigator or delegate, including sample and data forwarding process, to the project coordinator.

Activity 3: The project coordinator will finalize project activities for site teacher and student reference during the project year.

Objective 4: The project coordinator will facilitate Youth Area Watch project orientation and follow-up training for site teachers.

Activity 1: The project coordinator will prepare site teacher training material.

Activity 2: The project coordinator will invite principal investigators of participating projects to assist in the training and follow-up sessions.

Activity 3: Based on school district and principal investigator schedules, the project coordinator will set dates for the training and follow-up sessions.

Activity 4: The project coordinator will facilitate the orientation session by the second month of the project year for site teacher to become familiar with the philosophy behind Youth Area Watch, as well as the research project activities to be conducted.

Activity 5: The project coordinator will facilitate a follow-up session at the end of the project year for site teachers to share information and identify strategies for improving student activities.

Objective 5: The project coordinator will complete the student project orientation. All participating students from the community sites will collectively meet aboard a research vessel for the Youth Area Watch introduction and preliminary activity participation.

Activity 1: The Youth Area Watch principal investigator will solicit three bids for vessel hiring to conduct the student orientation.

Activity 2: The Youth Area Watch principal investigator will identify a vessel to hire for conducting the student orientation.

Activity 3: The project coordinator will invite research project principal investigators to participate in the student orientation.

Activity 4: Once a commitment is obtained by at least one research project principal investigator, a date will be set for student orientation.

Activity 5: The Youth Area Watch principal investigator will coordinate travel arrangements for student participation in the orientation.

Activity 6: In cooperation with the research project principal investigator(s), the project coordinator will conduct the student orientation to Youth Area Watch goals, responsibilities and activities.

Objective 6: With the oversight of site teachers, students conduct fish monitoring activities aboard research vessels according to research protocol identified by principal investigators.

Activity 1: The project coordinator will work with Evelyn Brown (\320-E-T-U) to coordinate student participation in research cruises.

Activity 2: The Youth Area Watch principal investigator will coordinate cost sharing for vessel hiring where possible.

Activity 3: With either teacher or research supervision (last year, students conducted activities on behalf of the fish monitoring researcher due to prohibitive weather for travel to the vessel), students will set nets at identified locations.

Activity 4: Students will check nets according to set protocol.

Activity 5: Students will take fish measurements according to set protocol.

Activity 6: Student will take fin and scale samples according to set protocol.

Activity 7: Students will track and analyze the fish monitoring data collected during the project year.

Objective 7: Students will conduct oceanographic data collection in their local communities. Site teachers will oversee these activities.

Activity 1: Students will take a bi-weekly water conductivity, temperature and depth reading. The water will be tested for salinity during this measurement as well.

Activity 2: A weather station will be installed at each site under the supervision of the site teacher. Students will measure the wind speed and direction, air temperature and barometric pressure.

Activity 3: Data will be collected at each site and transmitted to the project coordinator periodically.

Objective 8: Students at each site will collect blue mussels for pristane/mussel analysis.

Activity 1: Students will tag and identify mussel bed characteristics during fall and winter months at their local sites.

Activity 2: Students will note predator/prey activity at the identified mussel bed sites monthly.

Activity 3: Students will collect mussels twice a month throughout the year, and more intensely according to principal investigator request during the spring months.

Activity 4: Student will label and cold storage mussels for transport to the Auke Bay laboratory in Juneau.

Activity 5: Students will send mussels directly to Auke Bay once an adequate collection has accumulated.

Activity 6: Student will count mussels in the beds according to set protocol.

Activity 7: Students will compile site data for transmission to the project coordinator.



Objective 9: Students will assist local hunters/ technicians collecting harbor seal biological samples.

Activity 1: Local hunters will facilitate a local orientation to identify community procedures for sample collection participation.

Activity 2: Students will analyze an available sample to become acquainted with what is taken and what to look for in a sample. Students collect various parts of the seal for analyzing, which include: skin, blubber, teeth, stomach skull, liver, heart, kidney. In addition, measurements and weight are taken for each animal.

Activity 3: Students will assist local hunters in harvesting the harbor seals for sampling.

Activity 4: Students at local sites will participate in taking samples from harvested seals.

Activity 5: Students will assist the hunter/ technician in preparing the sample for shipment to the harbor seal management principal investigator.

Objective 10: Each community site will conduct a local research/restoration project.

Activity 1: The site teachers and project coordinator will work with participating students to identify a local research/restoration project.

Activity 2: Site teachers will work with project PIs were appropriate to develop protocol for student participation.

Activity 3: Students will conduct local project activities according to protocol and timelines set out by site teachers.

Activity 4: Students will provide data/samples to project PIs according to protocol.

Objective 11: Youth Area Watch students will interact with research project principal investigators, gaining a greater understanding of the affects of the oil spill on the ecosystem.

Activity 1: Principal investigators will commit to working with students directly at least once during the project year.

Activity 2: Students will work beside principal investigators during field work.

Activity 3: Students will independently conduct activities set out by the principal investigators.

Activity 4: Students will draw conclusions from their independent work to be reported at the annual Science Review.

Activity 5: Students will work with Community Involvement (/052) local facilitators and community members to increase awareness of restoration activities and the status of the ecosystem.

### **C. Cooperating Agencies, Contracts, and Other Agency Assistance**

The Chugach School District serves as the administrative agency for Youth Area Watch through their contract with the Department of Fish and Game. The school district has shown that it is an effective link to the students and communities impacted by the oil spill. As the administrative entity, the Chugach School District will maintain memoranda of agreement with the Valdez School District, Cordova School District and Kenai Peninsula Borough School District for the current communities identified in FY 97.

The Chugach School District continues to work with the University of Alaska in an effort to provide credit for progressively responsible activities and research conducted by students participating in Youth Area Watch. The district views the University of Alaska system as an integral partner in a continuum of active ecosystem awareness and restoration. Through the Native Marine Sciences Program at the University of Alaska Fairbanks, students will have the opportunity to further their understanding of research and restoration activities, as well as explore personal goals that may lead to a career in this field.

The Chugach School District continues to work with the regional organizations Chugachmiut and Chugach Regional Resources Commission to coordinate and exchange community information with regard to restoration activities. As the coordinating agency for community involvement, Chugach Regional Resources Commission works with the youth through the local facilitators so that students may participate in research and restoration activities.

The Chugach School District relies heavily on the commitment and participation of cooperating school districts involved in the project. Site teachers dedicate their time to the goals of Youth Area Watch, serving as an in-kind contribution.

Since the inception of the project, significant contributions have been made and are identified in the budget. Contractors have provided discounted services, as in the case of vessel hiring. Nets for fish monitoring were contributed by the Department of Fish and Game. Cooperating agencies provide technical assistance, student supervision and support for project activities.

In keeping with its commitment to secure additional support for Youth Area Watch project activities, the Chugach School District received funds from the Alaska Conservation Foundation. The school district was also awarded a grant to provide vocational support for the Youth Area Watch students in their exploration and activity participation. Lastly, the school district continues to redirect district funds to offset the full cost of the project.

## **SCHEDULE**

### **A. Measurable Project Tasks for FY98 (October 1, 1997 - September 30, 1998)**

September 25 -October 10, 1997:	Students apply for project participation
October 10 - 17, 1997:	Students selected for participation
October 21 - 23, 1997:	Site teachers receive protocol training
October 25 - 29, 1997:	Students receive protocol training
November 1, 1997	Local research project identified
November 10, 1997	Local project protocol developed
November 1 - 7, 1997:	Sites prepare weather stations
November 1 - July 30, 1998:	Students participate in research activities
March 1, 1998	Project Coordinator sends data to PIs
June 1, 1998	Project Coordinator sends data to PIs
June 1, 1998	Students complete project reports for FY98

### **Ongoing Activities:**

October 97 -September 98:	Student bi-monthly collection of mussels
October 97 -September 98:	Student mussel bed monitoring
October 97 -September 98:	Students participate in oceanographic cruises
October 97 -September 98:	Student weather station monitoring (2 x/wk)

October 97 -September 98:	Students participate in fish monitoring cruises
October 97 -September 98:	Students collect harbor seal samples with local hunters
October 97 -September 98:	Students conduct local project activities
October 97 -September 98:	Students assist in documenting local TEK
October 97 - September 98:	PIs interact and exchange information with students

## **B. Project Milestones and Endpoints**

October 17,1997:	Students selected for participation
October 30,1997:	Protocol training complete
November 1,1997:	Students conduct project activities
March 1, 1998:	Data/samples to PIs
June 1, 1998:	Data/samples to PIs and reports
complete	
October 17,1998:	Students selected for participation
October 30,1998:	Protocol training complete
November 1,1998:	Students conduct project activities
March 1, 1999:	Data/samples to PIs
June 1, 1999:	Data/samples to PIs and reports
complete	
October 17,1999:	Students selected for participation
October 30,1999:	Protocol training complete
November 1,1999:	Students conduct project activities
March 1, 2000:	Data/samples to PIs
June 1, 2000:	Data/samples to PIs and reports
complete	
October 17,2000:	Students selected for participation
October 30,2000:	Protocol training complete
November 1,2000:	Students conduct project activities
March 1, 2001:	Data/samples to PIs
June 1, 2001:	Data/samples to PIs and reports
complete	
October 17,2001:	Students selected for participation
October 30,2001:	Protocol training complete

November 1, 2001:  
March 1, 2002:  
June 1, 2002:

Students conduct project activities  
Data/samples to PIs  
Data/samples to PIs and reports  
complete

### **C. Completion Date**

Objectives identified in the project design will continue to serve the guidelines for community involvement within the civil settlement throughout the life of the restoration effort. It is expected that the Youth Area Watch project will be completed upon termination of the restoration process.

### **PUBLICATIONS AND REPORTS**

During the FY 97 project year, a video was completed of Youth Area Watch training, project activities and interaction with research and restoration projects. This video will be used to publicize the objectives of the project, as well as serve as a model for youth-researcher interaction and knowledge exchange. The video will be shown at education seminars as a state-of-the-best practice for science learning.

Reports on Youth Area Watch will be submitted to peer-review journals during FY 98 as invited. No cost for this effort is requested.

### **PROFESSIONAL CONFERENCES**

While professional conferences may be attended by either the principal investigator or the project coordinator during FY 98, none are currently scheduled outside of the annual science review in Anchorage.

### **NORMAL AGENCY MANAGEMENT**

Youth Area Watch is not a normal science curriculum component for students in the Chugach School District. The project developed out of a concern for the necessary involvement of oil spill impacted communities in the restoration effort. As a vital key in the ecosystem restoration, youth are the link to long-term enhancement; with this premise, a project allowing student participation in research and restoration efforts evolved.

In FY 97, the Chugach School District extended its efforts to the Cordova School District, Kenai Peninsula Borough School District and Valdez School District.

Through memoranda of agreement, students from these additional schools district are afforded the opportunity to participate in this project. The four school districts commit many hours of teacher time, facilities and other resources to ensure that the project is a success.

## **COORDINATION AND INTEGRATION OF RESTORATION EFFORT**

Youth Area Watch relies on the participation of Trustee Council funded projects to maintain coordination with restoration efforts. Through the commitment of principal investigators, youth conduct research activities for participating projects. Students work both independently, as well as beside researchers during the project year. Costs are shared between project to allow for increased research vessel time and one-on-one interaction between students and the researchers.

Various agencies provide the necessary technical assistance and resources. Local community facilitators from Community Involvement (/052) work with students and serve as chaperones for project activities. The education staff of the Prince William Sound Science Center provides provided technical assistance and SEA project coordination. School districts provide teacher time and facility space for activities.

A variety of funding sources and project contributions provide for the success of the project. In FY 97, the Chugach School District sought and received \$2,500 for the project from the Alaska Conservation Foundation; the school district also received \$100,000 to provide a continuum of education and career preparation for students participating in Youth Area Watch. School districts contribute \$40,000 in teacher time and \$20,000 in facility resources. The Prince William Sound Science Center provides \$25,000 in staff time. Communities and school districts contribute \$8,000 in lodging. Equipment in-kind contributions total \$7,000.

## **EXPLANATION OF CHANGES IN CONTINUING PROJECTS**

Student participation will increase in FY 98 to twenty-eight youth. In addition, participating students will introduce their science classes to the activities that they have conducted, broadening the impact of Youth Area Watch objectives. Site teachers will incorporate project activities into normal science classroom work.

Students will identify local restoration projects that they can participate in or develop at the community level. Under the direction of the site teacher, students will work with principal investigators and researchers to outline protocol for

their local project activities. This objective will allow for greater ownership of restoration efforts, as well as the possibility of extended activities into the summer months.

The Chugach School District will expand the scope of Youth Area Watch to include activities leading to career development among participating students. The district plans to foster the student growth and provide the framework for continuing the scientific learning process through additional grant sources.

#### **PROPOSED PRINCIPAL INVESTIGATOR**

Roger Sampson  
Chugach School District  
165 E. 56<sup>th</sup> Ave., Suite D  
Anchorage, AK 99518  
Office: (907) 561-3666  
Fax: (907) 561-8659

**1998 EXXON VALDEZ TRUSTE E COUNCIL PROJECT BUDGET**  
October 1, 1997 - September 30, 1998

Budget Category:	Authorized FY 1997	Proposed FY 1998						
Personnel		\$0.0						
Travel		\$0.0						
Contractual		\$140.4						
Commodities		\$0.0						
Equipment		\$0.0	LONG RANGE FUNDING REQUIREMENTS					
Subtotal	\$0.0	\$140.4		Estimated FY 1999	Estimated FY 2000	Estimated FY 2001	Estimated FY 2002	
General Administration		\$9.8						
Project Total	\$0.0	\$150.2		\$187.0	\$187.0	\$187.0	\$187.0	
Full-time Equivalents (FTE)		0.0						
Dollar amounts are shown in thousands of dollars.								
Other Resources								
Comments:								

**1998**

Project Number: 98210  
Project Title: Youth Area Watch  
Agency: Alaska Department of Fish and Game

FORM 3A  
TRUSTEE  
AGENCY  
SUMMARY



# 1998 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1997 - September 30, 1998

Personnel Costs:		GS/Range/ Step	Months Budgeted	Monthly Costs	Overtime	Proposed FY 1998
Name	Position Description					
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
Subtotal			0.0	0.0	0.0	
<b>Personnel Total</b>						<b>\$0.0</b>

Travel Costs:		Ticket Price	Round Trips	Total Days	Daily Per Diem	Proposed FY 1998
Description						
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
<b>Travel Total</b>						<b>\$0.0</b>

**1998**

Project Number: 98210  
 Project Title: Youth Area Watch  
 Agency: Alaska Department of Fish and Game

FORM 3B  
 Personnel  
 & Travel  
 DETAIL

October 1, 1997 - September 30, 1998

<p><b>1998</b></p>	<p>Project Number: 98210          Project Title: Youth Area Watch          Agency: Alaska Department of Fish and Game</p>	<p>FORM 3B          Contractual &amp;          Commodities          DETAIL</p>
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4/11/97

October 1, 1997 - September 30, 1998

## 1998

FORM 3B  
Equipment  
DETAIL

# 1998 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1997 - September 30, 1998

Budget Category:	Authorized FY 1997	Proposed FY 1998						
Personnel		\$60.0						
Travel		\$32.7						
Contractual		\$19.4						
Commodities		\$4.9						
Equipment		\$0.0	LONG RANGE FUNDING REQUIREMENTS					
Subtotal	\$0.0	\$117.0		Estimated FY 1999	Estimated FY 2000	Estimated FY 2001	Estimated FY 2002	
Indirect		\$23.4						
Project Total	\$0.0	\$140.4		\$175.0	\$175.0	\$175.0	\$175.0	
Full-time Equivalents (FTE)		1.0						
Dollar amounts are shown in thousands of dollars.								
Other Resources		\$212.5		\$212.5	\$212.5	\$212.5	\$212.5	
<p>Comments: <b>Personnel</b> - The Project Coordinator position has been increased to a full-time position, given the increase in the number of projects and planned activities associated with Youth Area Watch, an increase in students and project development responsibilities. <b>Travel</b> - Most student transport will be by charter between communities or to research site/cruises. Other student travel to Anchorage for the Science Review will be a project contribution. Only transport expenses are requested for project activities. All per diem expenses are a contribution to the project.</p> <p><b>Contractual</b> - An eighty-foot vessel will be hired for training and research cruises. Hiring of vessels will be coordinated with research PI cruises to maximize the length and number of trips. PWSSC staff provide technical assistance regarding protocol/protocol adherence. <b>Commodities</b> - Student equipment will only be purchase for 10 additional students. Each classroom site is allocated \$550 for student activities pertaining to Youth Area Watch.</p> <p><b>Indirect</b> - Administrative costs are calculated at 20% to account for the direct oversight of fiscal reporting and associated support at the school district administrative offices in Anchorage. In addition, costs have been included to offset the expenses that sites incur including, telephone, fax, postage and general support.</p> <p><b>Other Resources</b> - Teacher time (\$40,000), participating PIs (\$6,000), Youth Area Watch PI (\$10,000), PWSSC education staff (\$25,000), Facility space (\$9,000), equipment (\$20,000) and other grant funds (\$102,500).</p>								

1998

Project Number: 98210  
Project Title: Youth Area Watch  
Name: Chugach School District

FORM 4A  
Non-Trustee  
SUMMARY

## October 1, 1997 - September 30, 1998

Project Number: 98210  
Project Title: Youth Area Watch  
Name: Chugach School District

4/11/97

# 1998 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1997 - September 30, 1998

<b>Contractual Costs:</b>		Proposed
Description		FY 1998
A vessel up to 80 feet long will be hired for research and restoration training and cruises with participating projects. Students will come together for initial protocol training on a research vessel to get an overall orientation of their role in the ecosystem assessment and restoration. In addition, Youth Area Watch project activities will coordinate with research cruises to share vessel costs. It is estimated that eight vessel days will be hired at \$1,800 per day, totaling \$14,400.		14.4
The Prince William Sound Science Center provides technical assistance to Youth Area Watch during the project year. The staff of PWSSC assist with protocol development and training, as well as coordinating student participation in SEA project activities. Technical assistance is calculated at \$125 per hour for up to 40 hours during the project year, totaling \$5,000.		5.0
<b>Contractual Total</b>		<b>\$19.4</b>
<b>Commodities Costs:</b>		Proposed
Description		FY 1998
Personal gear for 10 new students including rain gear, shovels, boots and related supplies. Costs are calculated at \$100 per student, totaling \$1,000.		1.0
Supplies for each classroom site are necessary for group activities throughout the project year. Supplies will include water testing chemicals, sampling containers (beakers, plastic bags), water resistant note pads and general office supplies. Each classroom site is calculated at \$550 for seven sites, totaling \$3,850.		3.9
<b>Commodities Total</b>		<b>\$4.9</b>

**1998**

Project Number: 98210  
Project Title: Youth Area Watch  
Name: Chugach School District

FORM 4B  
Contractual &  
Commodities  
DETAIL

# 1998 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1997 - September 30, 1998

New Equipment Purchases:		Number of Units	Unit Price	Proposed FY 1998
Description				
				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 a <b>New Equipment Total</b>				\$0.0
Existing Equipment Usage:		Number of Units		
Description				
Laptop Computer		2		
Weather Stations		4		
Stereo Microscopes		4		
Video Recorder		1		
GPS		1		

**1998**

Project Number: 98210  
Project Title: Youth Area Watch  
Name: Chugach School District

FORM 4B  
Equipment  
DETAIL





## **EASTERN PWS WILDSTOCK SALMON HABITAT RESTORATION**

Project Number: 98220

Restoration Category: General Restoration

Proposer: Native Village of Eyak, USFS

Lead Trustee Agency: USFS

Duration: 3 Years

Cost FY 97: \$115,000

Cost FY 98: \$11,900

Geographic Area: Eastern Prince William Sound

Injured Resource/Service: Replacement of Lost Subsistence Services

**RECEIVED**  
APR 15 1997

**EXXON VALDEZ OIL SPILL  
TRUSTEE COUNCIL**

### **ABSTRACT**

The proposal for FY1998 is a continuation of project 97220. It consists of the monitoring of the instream habitat structures built in FY 1997, an analysis of the utilization of the structures by juvenile fish, an escapement count of coho salmon in October 1997, and a closeout of the project with a final report by September 1998.

## **INTRODUCTION**

### **NEED FOR THE PROJECT**

#### **A. Statement of Problem**

Levels of subsistence harvest have gradually increased in all of the spill area communities. However, subsistence harvests in Prince William Sound remain below pre-spill levels and, in some areas, the composition of the subsistence harvest has changed significantly. Subsistence users also report that the effort necessary to harvest resources has increased, and they continue to voice concerns about food safety.

Subsistence will have recovered when injured subsistence resources are healthy and productive and exist at pre-spill levels and people are confident that the resources are safe to eat. This project will attempt to replace injured subsistence services by enhancing salmon resources important to the Native Village of Eyak. Production of additional salmon through habitat improvement will provide additional subsistence opportunities and contribute to the overall restoration of subsistence resources in Prince William Sound.

#### **B. Rationale**

This project will directly contribute to the subsistence recovery objective as identified in the *Exxon Valdez* Oil Spill Restoration Plan. This project will target habitat enhancement of local salmon stocks that are utilized as a subsistence resource by the Native Village of Eyak. Habitat enhancement or restoration will increase the capability of local streams to produce additional salmon, and therefore provide increased subsistence resources and opportunities.

#### **C. Location**

The habitat enhancement work will be located in Plateau Creek in the Port Gravina area in eastern Prince William Sound. It will primarily benefit the communities of Cordova and Tatitlek.

## **COMMUNITY INVOLVEMENT**

One of the goals in this restoration effort is the direct involvement of the community, and especially the Native Village of Eyak in all aspects of the project. At the time this proposal is being written, the hiring of the 1997 field crews has not been completed, but we are striving to hire members of the local community for this year's work and in 1998.

## **PROJECT DESIGN**

### **A. Objectives**

1. Improve salmon spawning and rearing habitat conditions in four eastern PWS streams through the installation of log and boulder structures.
2. Educate student interns in the concepts and application of fisheries habitat management and incorporate their knowledge of local conditions and habitat in the habitat assessment.
3. Involve subsistence users from the Native Village of Eyak to the maximum extent possible.
4. Develop a baseline of information on existing wildstock salmon habitat conditions within the project area.

### **B. Methods**

In 1996, 11 streams were surveyed to determine the habitat available for coho salmon and the factors limiting production. The analysis of the data showed that unchangeable conditions, such as steep gradients or high flows, limited production in most systems. It was found, however, that winter habitat probably limits production in Plateau Creek, and this system provides the best opportunity for habitat enhancement. Fifteen to 20 habitat structures are planned to be built in FY1997, which will increase winter habitat.

In October 1997 (FY 1998) we will conduct adult escapement counts for baseline data. (Counts in 1996 were too late in the year for the peak escapement.) In 1998, the structures will be monitored to see how well they have withstood high flows, the amount of habitat created, and the utilization by juvenile coho salmon. Since observation of the utilization of the structures in winter will not be practical (the habitat will be covered with ice and the fish will not be active), mark and recapture population estimates of the enhanced habitat areas will be made in early spring before smolt migration. A final report will be written by the end of FY1998.

The first returning adults which may have benefitted from the efforts will not return until 1999. The Cordova Ranger District will conduct escapement counts and update the final report. No EVOS funding will be required for this.

### **C. Contracts and Other Agency Assistance**

No contracts or assistance from agencies other than the U.S. Forest Service will be required.

## **SCHEDULE**

### **A. Measurable Project Tasks for FY 98**

October 1 - 31, 1997	Conduct adult escapement counts at Plateau Creek.
April 15 - May 15, 1998	Dates dependent on snow levels and access. Conduct population estimates in enhanced areas.
June - July	Assess effects of spring runoff on structures. Repair if needed.
September	Complete final report.

### **B. Project Milestones and Endpoints**

Objectives 1 and 4, the habitat enhancement and baseline data collection will be completed in FY 1997. Involvement of local youth (objective 2) will continue in FY 1998 with the project monitoring, as will the close involvement with the Village of Eyak (objective 3). The project will proceed as outlined in the previous section, with closeout in September 1998. Adult escapement counts will be conducted by the Cordova Ranger District in 1999, but will not require EVOS funding.

### **C. Completion Date**

The project will be completed September 1998.

## **COORDINATION AND INTEGRATION OF RESTORATION EFFORT**

This project proposal has been closely coordinated with the fisheries staff of the Cordova Ranger District, USDA Forest Service. The project will use restoration techniques similar to those that were used by the Forest Service with the Montague Island Chum Salmon Restoration project (94139). Results from the Forest Service monitoring efforts on Montague Island will be incorporated into the habitat improvement prescriptions. Equipment from the ongoing Montague Island project and the Cordova Ranger District will be available for this project.

## **PROPOSED PRINCIPAL INVESTIGATOR**

David Schmid  
USDA Forest Service, Cordova Ranger District  
P.O. Box 280, Cordova, AK 99574  
(907) 424-7661 (telephone)  
(907) 424-7214 (Fax)

### **Principal Investigator**

David Schmid is the program manager and a fisheries biologist for the Cordova Ranger District. He has a B.S. degree in resource management from the University of Wisconsin, Stevens Point. He worked on the Glacier Ranger District for four years as a fisheries technician and two years as a fisheries biologist. During this time he managed the fisheries program and oversaw the construction of several fish ladders and other fisheries habitat restoration and enhancement projects. Since 1990 he has been the program manager on the Cordova Ranger District.

Ken Hodges will be the crew leader for the project and will be responsible assessing the structures and their utilization, conducting the escapement surveys, and writing the final report.

**1998 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET**

October 1, 1997 - September 30, 1998

<b>Budget Category:</b>	<b>Authorized FY 1997</b>	<b>Proposed FY 1998</b>						
Personnel	\$50.5	\$9.6						
Travel	\$1.4	\$0.0						
Contractual	\$42.0	\$0.0						
Commodities	\$4.2	\$0.9						
Equipment	\$6.4	\$0.0	<b>LONG RANGE FUNDING REQUIREMENTS</b>					
Subtotal	\$104.5	\$10.5		<b>Estimated FY 1999</b>	<b>Estimated FY 2000</b>	<b>Estimated FY 2001</b>	<b>Estimated FY 2002</b>	
General Administration	\$10.5	\$1.4						
Project Total	\$115.0	\$11.9						
Full-time Equivalents (FTE)	1.6	0.2						
Dollar amounts are shown in thousands of dollars.								
Other Resources								
Comments: Close out project. 96220, 97220. Final report due 4/15/99.								

**1998**

Project Number: 98220  
 Project Title: Eastern PWS Wildstock Salmon Habitat Enhancement  
 Agency: US Forest Service

**FORM 3A  
 TRUSTEE  
 AGENCY  
 SUMMARY**

**1998 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET**  
 October 1, 1997 - September 30, 1998

Personnel Costs:		GS/Range/ Step	Months Budgeted	Monthly Costs	Overtime	Proposed FY 1998
Name	Position Description					
D.Schmid	Project Leader	GS-11	0.3	5.2		1.6
K.Hodges	Fish Biologist	GS-9	1.2	3.9		4.7
Vacant	Bio Tech	GS-7	0.7	3.4		2.4
Vacant	Bio Tech	GS-4	0.5	1.7		0.9
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
Subtotal			2.7	14.2	0.0	
Personnel Total						\$9.6
Travel Costs:		Ticket Price	Round Trips	Total Days	Daily Per Diem	Proposed FY 1998
Description						
EVOS Restoration Symposium, RT Cordova to Anchorage						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
Travel Total						\$0.0

**1998**

Project Number: 98220  
 Project Title: Eastern PWS Wildstock Salmon Habitat Enhancement  
 Agency: US Forest Service

FORM 3B  
 Personnel  
 & Travel  
 DETAIL

Prepared:

2 of 4

4/15/97

**October 1, 1997 - September 30, 1998**

1998

Project Number: 98220 Project Title: Eastern PWS Wildstock Salmon Habitat Enhancement Agency: US Forest Service
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FORM 3B  
Contractual &  
Commodities  
DETAIL

3 of 4

4/15/97



**October 1, 1997 - September 30, 1998**

4/15/97



Analysis, Integration and Publication of Pre- and Post-Spill Data Necessary to Understanding Damage to, and Response of, Sea Otters and the Nearshore Ecological Community Following the Exxon Valdez Oil Spill; Submitted Under the BAA No. 52ABNA700049

Project Number: 98223  
Restoration Category: Research and Monitoring

Proposer: Lisa M. Rotterman, Ph.D.  
Enhydra Research

Lead Trustee Agency:

Cooperating Agencies:

Alaska SeaLife Center:

Duration: FY 98, 1st year, 1-year project

Cost FY 98: \$66,700

Cost FY 99:

Cost FY 00:

Cost FY 01:

Cost FY 02:

Cost FY 03:

Geographic Area: No Fieldwork

Injured Resource/Service: Sea Otters and the Nearshore Ecological Community

RECEIVED  
APR 15 1997

EXXON VALDEZ OIL SPILL  
TRUSTEE COUNCIL

#### ABSTRACT

Extensive new analysis, integration, and publication of pre- and post-spill data on sea otter movements, rehabilitation, carcasses and habitat use, and of data from repeated pre- and post-oil multi-species marine mammal surveys, will be undertaken so as to: a) understand EVOS damage to marine mammals and related natural communities; b) evaluate sea otter population processes affecting recovery; c) evaluate future response and restoration strategies; and d) generate benchmarks of sea otter population status.

## INTRODUCTION

Sea otters were severely impacted by the T/V Exxon Valdez oil spill (EVOS) and could not yet have recovered fully from the impacts of the spill. Their current status relative to recovery is unclear, and they are listed as "not recovering" in current Trustee Council publications. This project would:

- 1) Result in significant advances in the level and availability of knowledge concerning persistent impacts of the EVOS on the survival of adult sea otters.
- 2) Result in significant advances in the level and availability of knowledge about factors related to the survival and reproduction of sea otters released from post-EVOS treatment centers, and hence, provide information crucial to the development of future oil spill response strategy along the west coast.
- 2) Provide key information regarding distribution and abundance of sea otters, harbor seals, sea lions, and other marine mammals within the spill zone immediately pre-oiling to serve benchmarks against which current status of the population relative to total recovery can be gauged.
- 3) Provide information regarding the distribution, abundance, and behavior of sea otters and other marine mammals over time as oil advanced on their habitat, and over time following oiling. This information is the best available for gauging the number of individuals of sea otters, and perhaps other marine mammal species, exposed to large amounts of fresh crude oil in Prince William Sound. As such, it provides information useful: a) to understanding persistent lethal and sublethal impacts of the spill; b) for understanding post-spill behavior; c) for identifying areas that served as refuges from the oil; and d) for providing information useful in formulating future spill response strategy.
- 4) Provide an estimate of sea otter short-term mortality in Prince William Sound that is independent of all previous estimates.
- 5) Provide information about sea otter movements and population structure critical to the evaluation of techniques and study design used to monitor population status and to identifying factors that may be hindering the recovery of sea otters and the nearshore ecological community.
- 6) Provide information about population structure needed to understand population processes, such as immigration and/or emigration affecting population recovery and to evaluating the total geographic area over which sea otter populations may have been impacted by the spill.

No new field work or data acquisition is proposed in this project. The investigators propose to:

a) undertake new and extensive summarization and analyses of pre- and post-spill data on sea otters and other marine mammals from studies conducted between 1984 and early 1992.

b) integrate unpublished pre-spill baseline data with post-spill data;

c) prepare papers for publication in the primary scientific literature, making the information available to all interested persons.

New and previously unexplored scientific issues will be examined. The new and significantly expanded analyses of the post-spill data and the analysis and integration of multi-year pre-spill data will result in a greatly improved and more complete understanding of the spill's impacts on sea otters and the factors impacting recovery. Products will produce information crucial to understanding the process of recovery of the nearshore ecological community, to evaluating alternative response and restoration strategies, and to evaluating monitoring and damage assessment study methodology. Culmination of the project will be the preparation of manuscripts, and the publication in the primary scientific literature of 5 papers, containing many new, and all previously unpublished, data and findings.

Pre-spill and post-spill data are from the investigators' studies between May 1984 and Nov. 30, 1991 in both Eastern Prince William Sound (EPWS) and western Prince William Sound (WPWS).

#### A. Relevant Background

No significant closeout funding was provided between 1993-1995 for any of the investigators spill studies as it was for all other comparable studies and only 3.5 months of salary was provided in 1992 (see below). Because of this, funding is now being requested to ensure that unique and valuable data be developed and the relevant findings made available through the peer-reviewed primary scientific literature.

Dr. C. Monnett and Dr. L. Rotterman, who will undertake this project, began studies of sea otter development, reproduction, survival, population structure, behavior, growth, body condition, and movements in 1984. They pioneered large scale studies, including radio-telemetry studies and growth studies, of sea otter pups and weanlings, conducting studies in both eastern and western PWS. In 1987, they initiated a comprehensive study of adult female reproduction, survival, body condition and movements. When the spill occurred, they were halfway through a study of 59 radio-instrumented sea otter females.

Early in the morning on the day of the spill, the proposers initiated multi-species marine mammal fixed-wing aerial surveys to document the distribution and abundance of all marine mammals in areas that might be impacted by the flow of oil from the tanker. These surveys were repeated in areas as the oil progressed, and the behavior of marine mammals to the oil as it invaded their habitat was observed. With assistance from other researchers, such as Dr. R. Garrott, these surveys were repeated

many times between the day of the spill and January 1990. They provide the best information available on the number of sea otters and other marine mammals exposed to oil within Prince William Sound, the movements of marine mammals relative to the oil, and the locations of habitats providing refuge from the oil. No funding was ever provided for data analyses, report writing, or manuscript preparation related to this study. A very preliminary summary of some of the aerial survey data from this study was made in the fall of 1989 in the midst of intensive, year-round and daily field activity.

The proposers were the Principal Investigators on key sea otter damage assessment studies. Between 1989 and Nov. 30, 1991, they conducted year-round intensive field studies, including studies of: 1) female reproduction, health, and survival (PI: C.M.); 2) post-weaning survival (PI: L.M.R.); 3) movement patterns (PI: C.M.); 4) determination of the fate of sea otters released from EVOS treatment centers (PI: C.M.); 5) blood chemistry and hematology of adult females and weanlings (PI: L.M.R.); and 6) pre- and post-spill aerial (and to a lesser extent, boat) marine mammal surveys in Prince William Sound (using funding from the Mineral's Management Service).

These post-spill studies, which were unprecedented in both scope and content, were highly successful resulting in the acquisition of key information on the spill's impacts on sea otters. Dr. Monnett and Dr. Rotterman captured hundreds of sea otters, radio-instrumented 100 adult females and 64 pups, monitored 45 radio-instrumented otters from the treatment centers, and collected samples and carcasses for studies of toxicology, pathology, and clinical blood studies.

A key finding from their post-spill studies was that the post-weaning survival of sea otters born into the spill area more than a year after the spill was very poor, and was significantly lower than their concurrent counterparts in the unoiled eastern Sound. This finding remains, to date, the most definitive evidence of persistent damage to sea otters from EVOS (see below for further discussion).

Additionally, they demonstrated that female reproductive and survival rates were normal in the spill region in 1990 and 1991, but that adult females in the spill region may have higher rates of liver dysfunction compared with their counterparts in the eastern sound.

Their study of treatment center otters showed that survival and pupping rates of the animals released from the treatment centers was very low and was followed by an increase in mortality in the recipient population.

Their study of movements confirmed that significant interchange was not occurring among oiled (in WPWS) and unoiled (in EPWS) sea otter study populations, a finding crucial to evaluation of all post-spill studies. These movement studies also produced data that provides insight into the recovery process by showing that

sea otters from EPWS were not emigrating to the west, and thus, would not be affecting recovery rates.

All funding for these studies, including all professional and technician salaries, ended without notice on Nov. 30, 1991, when invoices for payment were returned. After several months, and the loss of experienced support staff, 3.5 months of salary were provided to cover:

1) preparation of 5 draft and final reports summarizing over two and a half years of continuous, year-round field work; and

2) all activities necessary to end this massive study (e.g., inventory and clearing of warehouses and offices, relocation of staff, etc.).

The 3.5 months of salary was insufficient to permit full analysis of all post-spill data, to permit analyses or integration of pre-spill data, and, thus, to develop manuscripts for publication.

Despite the short time frame allotted for report preparation, the proposers reported the basic results, addressing all contracted objectives from these studies, in final unpublished reports in 1992.

The aforementioned reports were accepted by the contracting agency as final reports with only minor modification being requested and made. However, these reports were not finalized by the contracting agency until May 1995. No content modifications were requested of the investigators between the submission of the final version of reports in 1992 and the agency certification of documents as "Final Reports" in 1995. However, the stated objectives of the NRDA reports were very simple (e.g., did survival rates of weanlings in unoiled versus oiled habitat differ?).

The population studies were highly praised by key sea otter peer reviewers advising the Chief Scientist. One investigator wrote something to the extent that before the successful completion of these studies, no one would have thought them possible.

#### **B. Work Undertaken With FY 1997 Funding**

In August of 1996, funding was approved by the Trustee Council to permit one of the investigators to spend 1.5 months on each of 4 papers to undertake analyses and writing necessary, and to consult with an expert marine mammal biostatistician and population modeler (L. Eberhardt), to produce 4 (of 8 originally proposed) papers.

A contract to L.M. Rotterman for \$40,200 for this project was approved and activated on February 1, 1997. To date, the investigators have made the following progress towards completion of the goals of the FY 1997 project: 1) completion of a manuscript entitled "Health, development, and survival of sea otter pups and weanlings", formatted for publication in the ..

Journal of Mammalogy (referred to hereafter as the "pup/weanling paper"; 2) completion of all data analyses and partial development of writing of a manuscript entitled "Length-mass relationships in sea otters in Prince William Sound after the T/V Exxon Valdez oil spill, also written for submission to the Journal of Mammalogy (expected date of submission to the contracting agency is April 21, 1997, a week ahead of schedule; 3) Significant progress has been made on data analyses and writing related to paper "Survival and reproduction of female sea otters in Prince William Sound, AK after the T/V Exxon Valdez oil spill, a paper targeted for Marine Mammal Science, and submission to the contracting agency is expected to be ahead of schedule. Work on the final paper "Age-specific reproduction of female sea otters in Prince William Sound, AK has not yet begun. In development of this paper, the authors hope to work with L. Eberhardt, as proposed. We expect to have some data to Dr. Eberhardt in May. Completion of the paper is expected to be at least by the target date, but due to funding delays, at this time of an actual completion target in advance of the target date cannot be estimated.

Extensive new data and analyses are presented in the pup/weanling paper and the length/mass papers referred to above. Actual time devoted to completion was well in excess of that for which funding was requested or provided, as anticipated both by us and by peer reviewers.

For example, in the pup/weanling paper, data from multiple independent pre- and post-spill studies of hundreds of radioinstrumented females and their pups, of radio-instrumented pups and weanlings, and of tagged pups are analyzed and integrated. Data on birth timing, body condition, growth rates, health, dependency period, and survival are all presented. In the corresponding report, the only objective was to determine whether the survival of weanlings in the eastern sound differed from that observed in the western sound.

A brief summary of important new data and findings resulting from these efforts, which involved: extensive new data preparation, summarization and analysis; integration of pre- and post-spill data bases; integration of data from independent post-spill studies; and literature review; and manuscript preparation, are given briefly below:

The purpose of the pup/weanling paper was to compare and to discuss measures of, and factors related to, the survival, development, and health of young sea otters through the period of dependency and after permanent separation from their mothers, and in so doing, to gain information necessary to determine whether there were chronic impacts of the 1989 T/V EXXON VALDEZ oil spill (EVOS) on the local populations of sea otters. The individuals studied were born in Prince William Sound, on average, more than a year after the T/V Exxon Valdez discharged oil into the sound and were not even conceived when the spill occurred.

Findings presented in the paper confirm and strengthen the result



presented in the final report (prepared in 1992) that post-weaning mortality was significantly and unusually low, compared to both pre-spill and post-spill studies. Proportions of individuals surviving after weaning in WPWS after EVOS were significantly lower in 1990 than in each of 3 separate studies in EPWS, one conducted concurrently and 2 conducted before EVOS.

Data on the survival of pups and weanlings were integrated in this paper and show that very few of the pups born in 1990 could have survived to ever contribute to population recovery as breeders. Although survival of this cohort was normal in the spill zone while maternal care was provided, there are multiple forms of evidence that indicated they were not as healthy as their EPWS counterparts during the pup period and may have been compromised at the time of weaning. Some evidence suggests that growth rates were negatively impacted and dependency periods lengthened. Both before and after the spill, pups tended to be born later in the year in WPWS than in EPWS. Pups captured in WPWS during 1990 had less mass and shorter total body length than pups caught in EPWS, but body condition (i.e., weight corrected for length) was similar.

When all forms of evidence from this study are considered in concert with evidence (discussed below) from the length mass paper, and with findings from other investigators regarding the persistence and distribution of oil in the environment and in sea otter prey in the period between 1989 and 1991, a pattern emerges that suggests that multiple oil-related factors likely contributed to the extremely high post-weaning mortality that was documented.

In the paper on length-mass relationships, we present new findings that show that the mean mass, adjusted for total body length, of male and female sea otters inhabiting oil-affected regions of Prince William Sound after the T/V *Exxon Valdez* oil spill was significantly less than that of individuals captured in the same or adjacent habitat in western Prince William Sound approximately a decade earlier, and than that of individuals inhabiting unoiled habitat in eastern Prince William Sound between 1984-1990.

### C. Relationship to other projects

This project is relevant to the Nearshore Vertebrate project which seeks to monitor the recovery of sea otters and to determine factors that may be limiting recovery. It provides information on oil spill damage and benchmarks that are necessary for achieving an understanding of sea otter recovery and gauging population status relative to recovery. It provides information on population status and the movements of individuals critical to designing studies aimed at elucidating factors impacts recovery and to monitoring recovery. It provides information directly relevant to evaluating monitoring methodology and evaluating previous evidence regarding post-spill population status.

## NEED FOR THE PROJECT

### A. Statement of Problem

Sea otters were one of the species most heavily impacted by the Exxon Valdez oil spill. Their current status is unclear. Recent Trustee Council documents list sea otters as "not recovered".

This project will result in extensive new analyses, integration, and publication, of pre- and post-spill data on sea otters directly needed to:

- \* evaluate monitoring methodology and interpretation of data regarding post-spill population status
- \* design studies aimed at elucidating factors impacting recovery sea otters and the nearshore ecosystem;
- \* provide information regarding population structure necessary and critical to the understanding of population processes impacting recovery (e.g., immigration or emigration), evaluating study design, and modeling and understanding the likely course of recovery of sea otters affected by the oil spill;
- \* provide possible the best information available on the number of sea otters and other marine mammals directly exposed to crude oil from the EVOS, information that is valuable in understanding observed persistent impacts;
- \* provide information about the behavior of sea otters, harbor seals, sea lions, and other marine mammals as oil invaded their habitat, and on areas that acted as refuges, information that is valuable in formulating future response activities;
- \* provide information on immediate pre-oiling abundance and distribution of sea otters, harbor seals, and sea lions that can act as benchmarks against which to gauge recovery;
- \* provide information regarding changes in distribution, abundance, and oiling of sea otters and other marine mammals in the months following the spill, data that valuable for providing an estimate of mortality independent of those provided previously;
- \* evaluate past and formulate future oil spill response policy
- \* better understand the geographic extent of damage to sea otters from the spill, and hence, better understand the process of recovery

None of the data proposed to be analyzed, integrated and published here from the Principal Investigator's marine mammal studies between 1984 and 1991 are currently available in the primary scientific literature. Lack of development of existing post-spill and pre-spill data from the principal investigators' studies on sea otter movements, the reproduction and survival of treatment center otters, habitat use, carcasses, and distribution and abundance relative to oiling in Prince William Sound, AK and adjacent regions has greatly constrained the ability of interested scientists, spill residents, and others to achieve a full understanding of the spill's impacts on sea otters and on the ecosystems in which they play a vital part. It has hampered

the interpretation of related findings from other spill-related studies. It has hampered development of meaningful recovery models for sea otters, and resulted in some sea otter-related restoration activities proceeding without important benchmarks against which to gauge current population status relative to recovery. Frustration over the lack of funding for the investigators to undertake analyses necessary for publication and other issues has caused some experienced and knowledgeable scientists to stop participating in the restoration process.

Information about population structure in and near the spill zone is needed to achieve a full understanding of both the geographic area over which sea otter populations or subpopulations were impacted by the spill, to understand the population processes impacting recovery, and to interpret data on distribution and abundance over time, and to formulate meaningful population models of recovery.

Survey data to be made available through this project can provide the best information available about the number of sea otters and other marine mammals exposed to the crude oil plumes that coursed through Prince William Sound in 1989. They also can provide information about the behavior of animals as oil invaded their habitat and about the locations of refuges from oiling. They provide immediate pre-oiling benchmarks for key areas within the sound against which to gauge current population status relative to recovery.

Information available in a 6-year pre-spill database is critical for the evaluation and interpretation of data on the number and ages of sea otter carcasses found during beachwalks. These beachwalk data have previously been interpreted as indicating an unusually high adult mortality in the spill zone in the years following the spill, and the data proposed to be developed here are critical in evaluation of that claim. Lack of funding to develop a 6-year pre-spill database has, thus, hampered understanding of current population status.

Information about factors affecting sea otters treated and released from post-EVOS treatment centers is critical to the formulation of future oil spill response policy and to evaluating the full extent of damage to sea otters from the Exxon Valdez oil spill.

This project will aid restoration by providing detailed data on EVOS damage to sea otters, without which restoration planning is hobbled. It will also aid restoration by providing benchmarks useful for assessing population status relative to recovery.

As noted in the background section, funding has never been provided to permit more than the most basic analyses of the relevant post-spill data necessary to address simple litigation-driven objectives.

Key peer reviewers have consistently recommended that these investigators be provided support to enable them to fully develop

their findings and to make them available to the general scientific community via publication in the primary literature. In 1991, for example, the key sea otter peer reviewer wrote "A complete analysis of the data from instrumented otters in Prince William Sound would certainly be a worthy objective and hopefully the Fish and Wildlife Service will be able to see that funding is available so that a complete analysis is carried out" (D. Siniff in a letter to C. Gorbics, Nov. 14, 1991).

Funds have also not previously been provided to permit the Principal Investigators to undertake the considerable effort to integrate the 1984-1989 (pre-spill) baseline data on the topics proposed here with post-spill data. The pre-spill study was interrupted by the grounding of the T/V *Exxon Valdez* on March 24, 1989 and funds were never provided to develop or to publish results of studies initiated in 1987.

Thus, funding is needed to permit the Principal Investigators the time needed to undertake the considerable effort required to finish analyses, integration, and writing required to ensure that the full value of the data and findings from this considerable and unique body of work is realized and available to the public and scientific communities. Peer reviewers to Trustee Council staff have repeatedly recommended that such funding be made available. This proposal seeks a relatively small amount of funding to undertake extensive data preparation, data analyses, manuscript preparation and publication.

#### B. Rationale/Link to Restoration

As noted throughout this proposal, information to be published as a result of the proposed work will provide benchmarks against which population status relative to recovery can be gauged. They will provide information valuable to planning future response efforts. They also provide baseline data that can serve as benchmarks of normalcy against which to evaluate whether further damage is occurring.

Due to lack of support to enable further data summarization and analyses, including the integration of a massive amount of baseline data from studies conducted prior to the spill, the scientific community and the public have not been able to fully evaluate and to understand the impacts of the spill on sea otters. Some publications have reported that there essentially was little relevant baseline, yet many years of comparable data exist on the key pieces of information necessary to evaluate damage and to monitor recovery.

Without reliable and detailed data on the population structure it is not possible to form conclusions about recovery status or to understand mechanisms impacting recovery.

#### C. Location

This project will be undertaken from the Principal Investigator's

offices in Kaneohe, HI, and in Homer, Alaska. The project's benefits will be realized globally, as information will be published in the primary literature and, thus, will be available to all interested persons.

## COMMUNITY INVOLVEMENT AND TRADITIONAL KNOWLEDGE

The Principal Investigators strongly support greater involvement of spill-area residents in Trustee Council restoration activities. The Principal Investigators have been residents of the spill area since 1984. This project involves only the development, integration and publication of data from studies already conducted. No new field work is proposed here and no technician aid is requested. Since the primary objectives of the proposal are to develop, integrate and publish results from the Principal Investigators' long-term studies, they will undertake all data analyses and writing required for project completion. Thus, it is unclear how, or if, additional spill area residents, other than the Principal Investigators, could be involved in implementation. However, the Principal Investigators would be willing to coordinate with the Spill Area-Wide Coordinator for the Trustee-sponsored Community Involvement Project to enhance communication of research findings from these projects to local communities.

Both before and after the spill, the Principal Investigators have ensured that interested members of the Native community had access to the findings from their research. For example, in the past, the authors have provided summaries of findings from their studies orally at meetings of the Alaska Sea Otter Commission, and provided copies of research publications to local Native corporations and to the Alaska Sea Otter Commission. Communication of the results from this project could take similar forms, or could involve non-technical oral presentations to community groups or the dissemination of reprints to local libraries. At present, no funds are requested to permit such enhanced communication. If the project is funded and activities such as community visits or reprint dissemination is requested by the spill Area-Wide Coordinator, the budget will need to be adjusted accordingly.

## PROJECT DESIGN

### A. Objectives

The general objectives of this proposal are to summarize data, conduct extensive new analyses, and produce five manuscripts for publication containing findings from studies of pre- and post-spill studies of sea otters and other marine mammals in Prince William Sound and the Gulf of Alaska.

The objectives of the 5 papers, and the associated analysis activities, are described in the section entitled "publications

and reports". However, in order to facilitate evaluation of the objectives of the project as a whole, a brief listing of the specific objectives of the analyses to be undertaken and the structure of the papers to be written are given below by paper. The amount of funding requested for each major portion of the project, given as amount of time for which funding is requested, is provided at the end the description of activities related to each paper.

1) The objectives of the paper entitled "Persistence of beach-cast carcasses of radio-instrumented sea otters in Prince William Sound, AK" are to:

a) analyze, integrate, and present data from studies conducted in Prince William Sound from the spring of 1984 to the end of Nov. 1991 on the condition and persistence of beach-cast carcasses of radio-instrumented sea otters as a function of age, sex, location, and time of year;

b) to develop and make available data directly relevant to the use of carcass counts obtained from annual beach walks of selected beaches as an indicator of mortality, and, hence, to make available data useful in the design of long-term monitoring of oil spill impacts and recovery following EVOS and future oil spills; and

c) make available data directly needed to evaluate previous interpretations of findings from the aforementioned carcass studies as evidence of persistent elevated mortality in sea otter adults related to the Exxon Valdez oil spill.

Time for which funding is requested: 1.5 months.

2) The objectives of activities related to the paper entitled "Movements of weanling and adult female sea otters in Prince William Sound, Alaska, after the T/V Exxon Valdez oil spill" are to:

a) summarize movement data crucial to achieving a full understanding of the damages to the affected population(s) and the likely course of recovery of that population;

b) provide information necessary to evaluate the basic design of many of the sea otter damage assessment studies and for guiding other analyses (e.g., to evaluating the assumption that the location of capture is indicative of the general area of residence and that sea otters from groups compared in other studies (e.g., the weanling study) do not live in the same habitat at any time;

c) to understand on a finer scale the movements of individuals being studied relative to categorization of habitat with regard to degree of oiling in order to guide other types of analyses;

d) evaluate whether there is significant immigration or emigration of sea otters into, or from, the oil spill affected region in general and whether sea otters within the oil spill affected region appear to be preferentially using or avoiding the most heavily oiled areas, and in so doing to provide data necessary to understand and to be able to make predictions about the recovery of sea otters in the oil spill affected areas.

Time for which funding is requested: 1.5 months.

3) The objectives of activities related to the paper entitled "Survival and reproduction of sea otters released from treatment centers after the T/V Exxon Valdez oil spill" are to:

a) analyze unique data and present findings on the survival and the reproduction of sea otters that were captured in oiled habitat after the Exxon Valdez oil spill, treated at centers established in response to the spill, and released back into unoiled habitat in PWS in the summer of 1989

b) compare the survival and reproduction of the treated animals to similar data on the recipient population collected concurrently, and prior to the spill

c) to examine factors possibly related to survival (e.g., age, condition upon capture, movement after release, center where treated, etc.)

d) to present data and findings critical to the evaluation of the efficacy of sea otter response and rehabilitation strategies

e) to present information relevant to assessing the total damage to, and, hence, the full recovery of, sea otter populations from the Exxon Valdez oil spill.

Time for which funding is requested: 2.0 months.

4) The objectives of activities related to the paper entitled "Immediate pre-oiling and repeated post-oiling surveys of marine mammals in Prince William Sound following the T/V Exxon Valdez oil spill" are to:

a) determine, compare, and publish information on the pre- and post-spill (meaning, in this case, the period after the arrival of oil in an area, rather than the period after the spillage of oil from the tanker) distribution and abundance of marine mammals (sea otters, harbor seals, Steller sea lions, orcas, and other marine mammals) in areas of Prince William Sound using repetitive surveys conducted between March 24, 1989 and Jan. 1990;

b) summarize, analyze and publish data relevant to assessing and interpreting oil impacts on sea otters, harbor seals, Steller sea lions, orcas, and other marine mammals.

c) make available information about the behavior of sea otters and other marine mammals as crude oil invades their habitat through observations made during aircraft surveys, and in doing so, gain insight that will be useful in the interpretation of carcass data, and evaluation of the potential alternative methods and strategies of response.

d) summarize, analyze, and make available information that is critical to estimating the numbers of sea otters, harbor seals, and sea lions exposed to crude oil in Prince William Sound, and, hence to the interpretation of other data regarding potential EVOS impacts on marine mammals;

e) provide information regarding locations affording refuge from oil for sea otters following the EVOS, and, in so doing, provide information valuable in the formulation of response activities in the event of future oil spills in the sound or other sea otter habitat.

Time for which funding is requested: 2.5 months.

5) The objectives of activities related to the paper entitled "Post-weaning movements of male and female sea otters in Prince William Sound, AK" are to:

a) summarize, analyze and present data on the post-weaning movements of radio-instrumented sea otters studied between 1984 and 1991 in Prince William Sound, Alaska in order to provide information about the population structure of sea otters within the spill zone and adjacent areas

b) determine whether there are sex-specific patterns in post-weaning movements of sea otters including the distance travelled, and the type of habitat resided in, following weaning

c) provide information directly relevant to understanding the population processes affecting recovery in the spill zone, e.g., to determining whether significant immigration into or emigration from the spill zone occurs that might impact population recovery within the spill zone

d) provide information necessary for evaluating trends in sea otter distribution and abundance in the oil spill area over time, for understanding population dynamics over the course of recovery, and, also to creating meaningful models of post-EVOS recovery of sea otters

e) provide a measure of the probable source pool for recruitment of sea otters to the oil spill areas

f) provide insight into total geographic area over which sea otters were likely impacted by the spill.

Time for which funding is requested: 2.5 months.

## B. Methods

The purpose of this project is to conduct extensive new analysis of existing data, integrate data from pre- and post spill studies, and to produce and submit five manuscripts for publication in the primary literature. No new field work or data collection is proposed.

We emphasize that extensive data summarization, new analyses, new integration of findings and literature review, etc., will be undertaken and presented in these most of these manuscripts. Some of the data which will be summarized and analyzed here were collected during NRDA studies and much of it was not. While directly relevant to interpreting oil spill impacts on, and recovery of, marine mammals and associated ecosystems, most of the findings to be contained within the papers have not been previously summarized in NRDA final reports. Thus, the final products will, in scope and content, go far beyond the objectives and products of previously funded Trustee Council activities and, hence, provide information previously unavailable.

Where possible, papers will incorporate additional data from studies conducted in Prince William Sound by the Principal Investigator and primary collaborator during 1984-1989. Much of the data to be analyzed and presented in publishable papers is . .



- from both pre-spill and post-spill studies conducted by the authors.

We emphasize that his project is not simply production of publishable papers from NRDA reports, and, as such, it requires additional time for completion beyond the time necessary to make the transition between report and publishable paper. Significant data preparation, data summarization, data analyses, literature review, and manuscript preparation is required for most of these papers.

No new field work is proposed as part of this project.

The section on "Publications" summarizes the types of data to be analyzed, integrated and presented in each paper. Background on the basic methods that were employed in the collection of the data to be analyzed are also presented in the "Publications" section.

#### C. Cooperating Agencies, Contracts, and Other Agency Assistance

None

### SCHEDULE

#### A. Measurable Project Tasks for FY 98 (October 1, 1996-September 30, 1997)

Assuming a starting date of October 1, 1997, manuscripts having the following, or similar titles, will be submitted to the contracting agency for review by the Chief Scientist and selected peer reviewers on, or before, the dates indicated. Other key dates are listed below.

October 1, 1997:	Begin data summary, analyses, and literature review needed for first paper
November 30, 1997:	Submit paper "Persistence of beach-cast carcasses of radio-instrumented sea otters in Prince William Sound, AK" to contracting agency" for review by Chief Scientist
January 30, 1998:	Submit paper "Movements of Weanling and Adult Female Sea Otters in Prince William Sound, Alaska, After the T/V Exxon Valdez Oil Spill" for review by Chief Scientist
January 15-18:	Attend Annual Restoration Workshop
January 19-26:	Travel to and attend The World Marine Mammal Scientific Conference in Monaco
April 1:	Submit paper "Immediate pre-spill and repeated post-spill surveys of marine mammals in Prince William Sound following the T/V Exxon Valdez oil spill" for review by Chief Scientist
June 30:	Submit paper "Survival and reproduction of sea otters released from treatment centers after the T/V Exxon Valdez oil spill for review by

Chief Scientist  
\*Sept. 15, 1998: Submit paper "Post-weaning movements of male and female sea otters in Prince William Sound, AK" for review by Chief Scientist

If the genetic study of sea otters proposed by Dr. Rotterman is funded, we request that funding for production of manuscript # 5 be delayed until FY 1999.

#### B. Project Milestones and Endpoints

The project objectives will be met when each successive manuscript is submitted for publication in the primary literature.

#### C. Completion Date

The project will be completed when the last manuscript is submitted, on, or before, Sept. 15, 1998.

#### PUBLICATIONS AND REPORTS

Below, information is provided on the types of data, comparisons to be made, and the manuscripts to be developed and submitted for publication in FY 1998 under this proposal.

1) Title: Persistence of beach-cast carcasses of radio-instrumented sea otters in Prince William Sound, AK  
Targeted Journal: Marine Mammal Science

When Manuscript will be Submitted: By November 30, 1997

Subject and Relevance to Restoration:

Information obtained about the number and estimated age of sea otters found as beach cast carcasses on selected beaches have been collected over the past 20 years in Prince William Sound and have been used to draw conclusions about the rates of mortality of various age classes of sea otters following the Exxon Valdez oil spill.

The data to be developed and made available through publication in this paper are directly relevant to the interpretation of carcass counts obtained on annual beach walks of selected beaches, and their use as an indicator of mortality.

Data will be presented on the condition and persistence of beach-cast carcasses of radio-instrumented sea otters as a function of age, sex, location, and time of year. Data from studies conducted in Prince William Sound from the spring of 1984 to the end of Nov. 1991 will be analyzed, integrated, and presented.

2) Title: Movements of Weanling and Adult Female Sea Otters in Prince William Sound, Alaska, After the T/V Exxon Valdez Oil Spill.

Targeted Journal: Marine Mammal Science

When Manuscript will be Submitted: By January 30, 1998

Subject and Relevance to Restoration:

In order to provide movement data necessary to address the issues discussed below, we monitored the movements of radio-instrumented weanling and adult female sea otters between October, 1989, and November, 1991. We made observations on individuals inhabiting the area affected by the oil spill in western Prince William Sound (WPWS) and on individuals living in ostensibly unaffected habitat in eastern Prince William Sound (EPWS).

Final analyses of these data (final with respect to the following issues only) will be undertaken, needed literature review undertaken, and a manuscript for publication in the peer reviewed scientific literature will be prepared.

Given the great potential mobility of sea otters documented in other studies, movement data are needed to test assumptions about the reliability of capture or single point observation location as a reliable indicator of the region normally inhabited, and hence, about the discreteness of compared groups of animals.

Thus, findings from this paper are critical to evaluating the design of past, ongoing, and future studies aimed at monitoring the recovery of affected sea otters and the associated nearshore ecological community.

Relatedly, in order to understand how to analyze data **within** the oil spill zone, it is necessary to understand on a finer scale the movements of individuals being studied relative to categorization of habitat with regard to degree of oiling.

A second major reason that movement data are needed is to understand and to be able to make predictions about the recovery of sea otters in the oil spill affected areas. Thus, movement data are needed to evaluate whether there is significant immigration or emigration of sea otters into, or from, the oil spill affected region in general and whether sea otters within the oil spill affected region appear to be preferentially using or avoiding the most heavily oiled areas.

Findings from this paper will provide information on whether sea otters tend to avoid oiled habitat, and hence, provide information critical to the evaluation of data on distribution and abundance of sea otters over time in certain areas as an indicator of population recovery.

Finally, data to be reported herein will provide information needed to model recovery of sea otters in affected areas in

Prince William Sound by providing information about whether the recovery of the sea otter population in the oil spill affected region of Prince William Sound will likely be a direct function of the rates of survival and reproduction of the sea otters in the affected habitat or whether there is significant influence from emigration or immigration from or to the area.

3) Title: Immediate pre-oiling and repeated post-oiling surveys of marine mammals in Prince William Sound following the T/V Exxon Valdez oil spill.

Targeted Journal: Marine Mammal Science

When Manuscript will be Submitted: By April 1, 1998

Subject and Relevance to Restoration:

In this portion of the project, we will summarize, analyze, and publish data from surveys on the distribution, abundance and behavior of marine mammals in Prince William Sound both immediately prior to, immediately after, and for a number of months after, the arrival of oil from the T/V Exxon Valdez. These unique data are relevant to assessing and interpreting oil impacts on sea otters, harbor seals, Steller sea lions, orcas, and other marine mammals.

Objectives:

The objectives of this study are to summarize, analyze and publish data from fixed-wing and, to a much lesser extent, boat, surveys of marine mammals in selected areas of Prince William Sound conducted between the morning of March 24, 1989 through Jan. 5, 1990 in order to:

a) Determine and compare the pre- and post-spill (meaning, in this case, the period after the arrival of oil in an area, rather than the period after the spillage of oil from the tanker) distribution and abundance of sea otters and other marine mammals in areas of Prince William Sound using repetitive surveys;

b) make available information about the behavior of sea otters and other marine mammals as crude oil invaded their habitat through observations made during aircraft surveys, and in doing so, gain insight that will be useful in the interpretation of carcass data and evaluation of carcass data and evaluation of the potential alternative methods and strategies of response.

4) Title: Survival and reproduction of sea otters released from treatment centers after the T/V Exxon Valdez oil spill

Targeted Journal: Conservation Biology

When manuscript will be submitted: By June 30, 1998

Subject and Relevance to Restoration:

This paper will present findings on the survival and the reproduction of 45 sea otters that were captured in oiled habitat . .

after the Exxon Valdez oil spill, treated at centers established in response to the spill, and released back into unspoiled habitat in PWS in the summer of 1989. Data of the survival and reproduction of these animals will be compared to similar data on the recipient population collected concurrently, and prior to the spill. Factors possibly related to survival (e.g., age, condition upon capture, movement after release, center where treated, etc.) will be examined.

The data to be presented in this paper are the only existing data available for evaluating the long-term fate of sea otters released into their natural habitat following a large scale oil spill and subsequent rehabilitation effort. Hence, they are critical for the evaluation of the efficacy of the rehabilitation strategy and for the planning of future response policy. They also provide insight into the probable fate of the other \* sea otters released from the EVOS treatment centers back to the wild and so provide heretofore unconsidered information relevant to assessing the total damage to, and, hence, the full recovery of, sea otter populations from the Exxon Valdez oil spill.

5) Title: Post-weaning movements of male and female sea otters in Prince William Sound, AK

Targeted Journal: Canadian Journal of Zoology

When manuscript will be submitted: By Sept. 15, 1998

Subject and Relevance to Restoration:

Data on the post-weaning movements of radio-instrumented sea otters studied between 1984 and 1991 in Prince William Sound, Alaska, will be examined. The movements of male and female sea otter weanlings will be compared, including the distance travelled, and the type of habitat resided in, following weaning. Data on the movements of mother-pup pairs prior to weaning will also be summarized and compared to post-weaning movement patterns.

Information that will be published in this paper will provide direct information necessary for evaluating trends in sea otter distribution and abundance in the oil spill area over time and for understanding population dynamics over the course of recovery.

For example, it is directly relevant to the interpretation of mother-pup counts obtained to estimate population or area-wide pupping rates. Since available data indicate that weanlings are probably the age class of sea otters most vulnerable to the chronic impacts of the spill, this information can provide guidance to the design of sampling efforts aimed at assessing the impacts of the spill on the nearshore sea otter (and other predator) prey communities.

This paper will also provide information directly relevant to determining population structure (and interpreting data on

genetic structure), knowledge of which is required before meaningful interpretation can be made of trends in distribution and abundance over time.

It will also provide a measure of the probable source pool for recruitment of sea otters to the oil spill areas. Conversely, it may provide some insight into the probable magnitude of emigrations from the area.

#### PROFESSIONAL CONFERENCES

Funding for registration and travel expenses needed to permit the Principal Investigator to present a paper at, and to attend, one professional conference in FY 1998 is requested. No previous conference funding has ever been requested from, or provided to, the Principal Investigator to present findings from the extensive and long-term EVOS-related studies of these investigators.

The conference is The World Marine Mammal Scientific Conference, to be held January 20-25, 1998, in Monaco. This conference is a joint conference combining both the XII Biennial Conference for the Society for Marine Mammalogy and the 12th Annual Conference of the European Cetacean Society. The program for this conference is currently under development. The Principal Investigator would anticipate presenting a spoken paper presenting a summary of oil spill-related findings at this conference. No presentation of the findings from the long-term studies of these investigators have been made to the Marine Mammal Society, despite the obvious relevance to the research its members. Presentation at this conference would be particularly important because of the large European contingent expected to attend and the history of oil spills affecting marine mammals in Europe.

#### COORDINATION AND INTEGRATION OF RESTORATION EFFORT

This project is directly relevant to the Nearshore Vertebrate Ecosystem Project. Publication of these papers will provide information crucial for understanding current population status. It will provide benchmarks for population status in key areas.

This project will provide information key to evaluation and interpretation of certain monitoring techniques and study designs in the Nearshore Ecosystem Project.

This project is also directly relevant to the proposed project to conduct genetic analyses to assess population structure of sea otters within the spill zone. If the genetics study is funded, we request that funding for the last of activities proposed here be delayed until FY 1999, and adjustment made of due dates to permit maximum flexibility in work schedule.

Data on movements to be presented here complements and aids interpretation of findings from the genetics study.

**PROPOSED PRINCIPAL INVESTIGATOR**

Lisa M. Rotterman, Ph.D.  
Enhydra Research  
Temporary address:  
44140 Mui Pl. #8  
Kaneohe, HI 96744  
Telephone: 808-236-0771  
Fax: 808-236-0771

**PROPOSED PRIMARY COLLABORATOR**

Charles Monnett, Ph.D.  
Enhydra Research  
Temporary address:  
44140 Mui Pl. #8  
Kaneohe, HI 96744  
Telephone: 808-236-0771  
Fax: 808-236-0771

## PRINCIPAL INVESTIGATOR

Dr. Lisa M. Rotterman will be Principal Investigator on this project and Dr. Charles Monnett will be the Primary Collaborator. In addition to having extensive research experience in the spill region, the Principal Investigator is a spill area resident and has resided in the spill region since 1984.

Working in full collaboration, Dr. Monnett and Dr. Rotterman have conducted original research on sea otter population ecology, behavior, development, and genetics in Alaska since 1984. Their sea otter research has included, but not been limited to, research on sex-, age-, and locality-specific: survival rates and causes; reproductive patterns and rates; movement patterns; morphology; body condition; and growth. They have also conducted studies on sea otter population structure and molecular and population genetics. Their sea otter studies have been multi-year, year-round studies in which hundreds of radio-instrumented individuals are studied intensively. They have developed indices by which to assess and compare sea otter populations status generally, and under different resource regimes, specifically.

Dr. Monnett and Dr. Rotterman pioneered studies on sea otter pups and weanlings. At the time of the spill, and until their studies were taken over by government researchers in 1992, they were the only scientists in the world to have conducted large-scale growth and telemetry studies on these age classes. This work permitted the post-spill studies on weanling survival, which produced the most definitive evidence of chronic damage to sea otter populations from the spill to date.

Dr. Monnett and Dr. Rotterman hold the best, most comparable, and in many cases the only baseline data available on the growth, reproduction, body condition, survival and movements of sea otter females, pups and weanlings in Prince William Sound.

As discussed more below, Dr. Monnett and Dr. Rotterman collaboratively conducted much of the post-spill field research on sea otters until 1992. They conducted pre- and post oiling marine mammal surveys in WPWS in 1989, including surveys initiated on the day of the spill. They captured, instrumented and monitored approximately a hundred and sixty sea otters in order to evaluate the impacts of the spill on adult female and weanling survival, health, and movements, and on female reproduction. They collected hundreds of samples for toxicology, pathology, blood chemistry and other studies. They also successfully undertook studies to evaluate the efficacy of the post-spill sea otter rehabilitation program by monitoring the post-release fate of sea otters from the treatment centers.

Dr. Monnett and Dr. Rotterman have written over 30 reports and publications based on their sea otter research.

Individual information about the qualifications of the two researchers are provided below.



Dr. Rotterman was the Principal Investigator on two major facets of the post-spill sea otter studies: 1) studies aimed at determining the impact of the spill on weanling survival; and 2) the impacts of the spill on the health of adult female and weanling sea otters as assessed through evaluation of blood chemistry and hematology. She has a Ph.D. and a M.S. from the Department of Ecology and Behavioral Biology at the University of Minnesota and a B.S. from the University of Maryland in the field of Conservation and Resource Development, with speciality in Fish and Wildlife. The specialities of her Ph. D. program were population and community ecology, evolution, and behavior. She has a second area of Ph.D.-level expertise in the fields of population, quantitative, and molecular genetics and earned a minor in Genetics as part of her Ph.D. program.

The topic of her Ph.D. dissertation was the impacts of population fragmentation and reduction on genetic variability and structure within and among populations of sea otters, and the implications of current genetic status to long-term viability. The field portions of her doctoral research were undertaken in Alaska, particularly in Prince William Sound. She was twice appointed as a Guest Researcher in the Laboratory of Viral Carcinogenesis in the Genetics Section at the National Cancer Institute, National Institutes of Health where the laboratory portions of her doctoral research was undertaken.

In addition to her research on sea otter ecology and genetics, she has many years of experience conducting research in the fields of avian ecology and non-human primate toxicology and infant development. She has additional research experience on other marine mammals, and caribou.

#### **OTHER KEY PERSONNEL**

Dr. Monnett will be the Primary Collaborator on this project. Dr. Monnett was the Principal Investigator on several key portions of the post-spill sea otter studies: 1) studies aimed at evaluating the impact of the spill on female health, reproduction and survival; 2) studies of the movement patterns of sea otters after the spill; 3) studies aimed at determining the efficacy of the sea otter rehabilitation program; and 4) pre- and post-spill aerial (and to a lesser extent, boat) marine mammal surveys (sea otters, harbor seals, sea lions, and other marine mammals) in oiled and adjacent areas of PWS, which he initiated on the morning of the spill.

Dr. Monnett has a Ph.D. from the Department of Ecology and Behavioral Biology at the University of Minnesota and a B.S. from the University of Washington in Zoology. He also has training in the veterinary sciences and is a certified veterinary technologist. He holds a private pilot's license and is certified as a commercial diver.

The topic of Dr. Monnett's Ph.D. dissertation was "Patterns of Movement, postnatal development and mortality of sea otters in

Alaska" in which studies of sea otter pups and weanlings were pioneered.

In addition to his research on sea otters, he has many years of experience conducting research in the fields of avian ecology and non-human primate toxicology and infant development. He also has additional research experience on other marine mammals.

**1997 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET**

October 1, 1996 - September 30, 1997

Budget Category:	Authorized FFY 1977	Proposed FFY 1998						
Personnel		\$0.0						
Travel		\$4.7						
Contractual		\$55.0						
Commodities		\$1.0						
Equipment		\$0.0						
Subtotal	\$0.0	\$60.7	LONG RANGE FUNDING REQUIREMENTS					
Indirect		\$6.0	Estimated FFY 1999	Estimated FFY 2000	Estimated FFY 2001	Estimated FFY 2002	Estimated FFY 2003	
Project Total	\$0.0	\$66.7						
Full-time Equivalents (FTE)		0.0						
Dollar amounts are shown in thousands of dollars.								
Other Resources								
<b>Comments:</b>  Indirect costs = 10% of direct costs								

**1997**

Project Number: 94223  
 Project Title: Publication of Sea Otter Data  
 Name: Lisa M. Rotterman, Ph.D.

FORM 4A  
 Non-Trustee  
 SUMMARY

Prepared:

1 of 4

4/13/97

# 1997 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1996 - September 30, 1997

Personnel Costs:				Months Budgeted	Monthly Costs	Overtime	Proposed FFY 1998	
Name	Position Description							
							0.0	
							0.0	
							0.0	
							0.0	
							0.0	
							0.0	
							0.0	
							0.0	
							0.0	
							0.0	
							0.0	
							0.0	
							0.0	
Subtotal				0.0	0.0	0.0		
						Personnel Total	\$0.0	
Travel Costs:				Ticket Price	Round Trips	Total Days	Daily Per Diem	Proposed FFY 1998
Description								
TRAVEL TO ANNUAL MEETING HNL-ANC			0.5	1	3	0.2	1.1	
TRAVEL TO TECHNICAL SESSION HNL-ANC			0.5	1	2	0.2	0.9	
TRAVEL TO WORLD MARINE MAMMAL SCIENTIFIC CONF. IN MORACO			1.1	1	8	0.2	2.7	
							0.0	
							0.0	
							0.0	
							0.0	
							0.0	
							0.0	
							0.0	
							0.0	
							0.0	
							0.0	
Travel Total							\$4.7	

1997

Prepared:

2 of 4

Project Number:  
Project Title: Publication of Sea Otter Data  
Name: Lisa M. Rotterman, Ph.D.

FORM 4B  
Personnel  
& Travel  
DETAIL

4/13/97

**1997 EXXON VALDEZ TRUST COUNCIL PROJECT BUDGET**

October 1, 1996 - September 30, 1997

<b>Contractual Costs:</b>		<b>Proposed</b>
<b>Description</b>		<b>FFY 1998</b>
SALARY OF LISA ROTTERMAN, 10 MOS. @ \$5.0/MO.		50.0
PAGE COSTS FOR 5 PAPERS @ \$1.0		5.0
<b>Contractual Total</b>		<b>\$55.0</b>
<b>Commodities Costs:</b>		<b>Proposed</b>
<b>Description</b>		<b>FFY 1998</b>
SUPPLIES		1.0
<b>Commodities Total</b>		<b>\$1.0</b>

**1997**

Prepared:

3 of 4

Project Number:  
Project Title: Publication of Sea Otter Data  
Name: Lisa M. Rotterman, Ph.D

FORM 4B  
Contractual &  
Commodities  
DETAIL

4/13/97

October 1, 1996 - September 30, 1997

1997

Project Number: \_\_\_\_\_  
 Project Title: \_\_\_\_\_  
 Name: \_\_\_\_\_

## FORM 4B Equipment DETAIL



## **Project Title: Port Graham Pink Salmon Subsistence Project**

Project Number: 98225  
Restoration Category: General Restoration  
Proposer: Port Graham IRA Council  
Lead Trustee Agency: ADF&G  
Cooperating Agencies: Port Graham IRA Council  
Alaska SeaLife Center  
Duration: 3<sup>rd</sup> year, 5 year project  
Cost FY 98 \$76,500  
Cost FY 99 \$77,200  
Cost FY 00 \$78,500  
Geographic Area: Port Graham, lower Cook Inlet  
Injured Resource/Service: Pink Salmon/Subsistence

**RECEIVED**  
APR 11 1997

**EXXON VALDEZ OIL SPILL  
TRUSTEE COUNCIL**

### **ABSTRACT**

This project will help supply pink salmon for subsistence use in the Port Graham area during the broodstock development phase of the Port Graham hatchery. Because local runs of coho and sockeye salmon, the more traditional salmon subsistence resource, are at low levels pink salmon are being heavily relied on for subsistence. This project will help ensure that pink salmon remain available for subsistence use until the more traditional species are rejuvenated. Two strategies are being employed; increased fisheries management surveillance to maximize use of adult pink salmon return and increasing marine survival of hatchery produced pink salmon.



## **Introduction**

This project will help underwrite the hatchery production of pink salmon for subsistence use in Port Graham. Normally pink salmon are not heavily utilized for subsistence. However, the local sockeye run has been very depressed and is just now beginning to respond to rehabilitation efforts, and the coho subsistence harvest is at about 15% of its historic level. This has resulted in a sharp increase in the number of pink salmon harvested for subsistence in recent years. Unfortunately, the pink run to Port Graham is also suffering. Escapement into the Port Graham River has barely met the minimum goal for four of the last five years (the 1995 return was somewhat better).

A salmon hatchery is being developed in Port Graham. Its principal mission is to build the pink salmon run back up to levels that will allow commercial exploitation. When this objective is achieved the impact of the subsistence harvest on pinks will be negligible. At this point in time however, the subsistence harvest has a significant impact. The hatchery is in the broodstock development phase. The more eggs that are put in incubation the faster the hatchery will achieve its goals. The low pink returns to the Port Graham River coupled with the subsistence harvest on the hatchery returns is limiting the number of eggs that can be put in the hatchery and extending the time it will take for the hatchery to build the broodstock it needs to become self sufficient.

The EVOS clean-up effort had a negative impact on the Port Graham pink salmon as it did on the local coho and sockeye runs. Boom deployment during the early phases of the clean-up trapped a large number of outmigrating pink salmon fry in the boom curtain on the ebbing tides causing high levels of mortality. It is possible that these losses are contributing to the poor even year returns that have been experienced recently.

This project is a small piece of the overall Port Graham pink salmon enhancement program. It comprises about a third of the overall Port Graham pink salmon enhancement budget. Port Graham pink salmon enhancement program complies with all state policies governing salmon enhancement activities including disease, genetics and harvest management. All required reviews and permits have been obtained for the hatchery program including this project. This project is designed to become self-sustaining beyond the development stage which is currently estimated to occur by the end of the decade.

## **NEED FOR PROJECT**

### **A. Statement of Problem**

The salmon runs to the Port Graham area are at low levels, partly as a result of the *Exxon Valdez* oil spill. As a consequence it has become more difficult for Port Graham villagers to meet their subsistence needs for salmon. Because of their four to five year life cycles, it will take a long time for the sockeye and coho runs to rebuild. A large number of the pink salmon that are being produced by the hatchery now being developed in Port Graham are being taken in the local subsistence fishery. Although the subsistence harvest of hatchery fish is helping to make up for

the lack of wild fish, it is making it far more difficult for the hatchery to develop the broodstock it needs to become self sufficient. Unless the schedule for developing broodstock can be maintained, the hatchery will lose its positive benefit/cost ratio and may have to be closed.

It is appropriate that the hatchery contribute pinks to the subsistence fishery. However, extraordinary methods will need to be employed for the hatchery to provide for the subsistence fishery as well as maintain its broodstock development schedule. These will include procedures to enhance the survival of juvenile pinks released from the hatchery, and coordinating with ADF&G to maximize the number of wild adult pink salmon returning to Port Graham that can be collected for broodstock.

#### **B. Rationale/Link to Restoration**

The importance of subsistence to the Native villages in the oil spill area has been recognized by the EVOS Trustee Council in its November 1994, *Exxon Valdez Oil Spill Restoration Plan*. This project will help preserve the subsistence lifestyle in Port Graham by providing additional salmon for subsistence needs. Harvest of these hatchery produced salmon will take pressure off the local wild runs; helping them in their recovery effort. Using an enhanced resource to replace harvest of an injured resource is an accepted strategy under the Restoration Plan.

#### **C. Location**

The project will be conducted at Port Graham with the bulk of the benefits accruing to the Port Graham village.

### **COMMUNITY INVOLVEMENT**

This proposal is being submitted by the Port Graham Village Council. The Port Graham hatchery is owned and operated by Port Graham Hatchery, Inc., an arm of the Port Graham IRA Council. The Port Graham Village Council will manage this project under a contract with ADF&G.

### **PROJECT DESIGN**

#### **A. Objectives**

Use the Port Graham hatchery to provide pink salmon for local subsistence use while maintaining the hatchery's pink salmon broodstock development schedule.

#### **B. Methods**

This will be the third year of a proposed five year project. Two basic strategies will continue to be employed to meet the objective. The first will be to supplement the ADF&G monitoring of

the Port Graham pink salmon return and the second will be to enhance the juvenile to adult survival of the hatchery produced pink salmon through an extended rearing program. A brief discussion of each approach is given below.

The Port Graham River pink salmon run is the source of the hatchery broodstock. A program has been established to work closely with ADF&G in monitoring the pink salmon return to Port Graham each year in order to get as precise an estimate as possible on the wild and hatchery return. This program supplements the normal management stream and bay surveys of Port Graham that ADF&G conducts. It includes additional stream surveys and closely monitoring the subsistence fishery harvest. This program has established regular lines of communications between Port Graham and ADF&G. By coordinating effort and keeping close track of the pink salmon return, it has been possible to maximize the harvest of pink salmon while ensuring that the Port Graham river pink salmon escapement goal is met. This program will be continued in FY 98.

The second approach will apply techniques to increase the fry to adult (marine) survival of hatchery produced pink salmon. Normal hatchery practice involves holding pink salmon fry in saltwater pens after they emerge from the incubators. These fish are put on feed and held until the first mature zooplankton bloom which usually occurs in the later part of May in the Port Graham area. Normal holding time is 3 to 4 weeks. The marine survival with this technique has been poor; ranging between 1 and 2.5%.

Test lots of pink salmon fry reared at Port Graham to an average weight of 8 grams (the threshold size at which pinks leave the near shore area for the high seas) had survival rates of 7% to 10%. Although this was very encouraging there are major problems with holding pink salmon fry the four months it takes to rear them to 8 grams. First, rearing fish to that size is expensive. Second, there is a high risk that fish held that long may contact disease or otherwise be injured or killed. Of particular concern is the potential for the rearing fish to contact "warm water vibrio", a highly contagious bacterial infection that pink salmon fry are susceptible to if reared in salt water warmer than 10° C. A group of pink salmon fry that were intended to be reared to an average weight of 8 grams under this project in FY 96 had to be released early because of an outbreak of warm water vibrio.

Studies undertaken at other pink salmon hatchery facilities in the state indicate that rearing salmon to a minimum of one gram also greatly enhances marine survival. Nearly eight times as many fry can be reared to 1 gram rather than 8 grams for the same cost. In addition, the reduced holding time required for producing 1 gram fish as opposed to 8 grams reduces the risk of loss from injury or disease. A group of pink salmon fry were successfully reared to the 1 gram size and released as part of this project's FY 96 activities. Unless analysis of the marine survival of the FY 96 group (to be completed by the fall of 1997) indicates that this strategy is not working, a similar, perhaps larger, group of 1 gram fry will be produced in FY 98.

The Port Graham hatchery now has the capability to produce modest amounts of heated water, both fresh and salt. This provides the potential to accelerate development and growth of small groups of fish. In FY 97 a lot of 20,000 pink salmon is being incubated and reared on heated

water with the objective of achieving a minimum average weight of 1 gram in time for release into the mature zooplankton bloom in late May. A search of the literature and conversations with other pink salmon hatcheries in Alaska, indicate that a test of this sort has never been conducted. However, it would seem that releasing large size pink salmon fingerling into the mature zooplankton bloom would greatly enhance marine survival. If the project is successful in releasing a lot of 1 gram fingerling into the mature zooplankton bloom in FY 97, it will be repeated in FY 98 with a group containing 250,000 fry.

All fish in both the 1 gram fingerling lot reared in ambient temperature water and the 1 gram fingerling lot produced with heated water will be otolith marked with a separate mark for each lot. For comparison purposes a third lot of pink salmon will receive the normal treatment of incubating and rearing in ambient temperature water for release into the zooplankton bloom. This lot will not be marked.

**SUPPLEMENTATION CRITERIA.** This is a supplementation project. The following is a brief discussion of how the project fits under each of the supplementation criteria presented in the *Invitation to Submit Restoration Projects for Federal Fiscal Year 1996 and Draft Restoration Program: FY 96 and Beyond*, March 1995, pages 34-35.

**Benefits of Supplementation.** This project will provide additional pink salmon for harvest in the subsistence fishery in the Port Graham area. By shifting some of the subsistence harvest to hatchery salmon this project will help Port Graham wild salmon stocks recover from their present low levels.

**Generic Risk.** The Port Graham pink salmon hatchery program was reviewed by the ADF&G, CFMD Genetics Section who determined that the program (which includes this project) meets all criteria of the state Genetics Policy for Salmon Enhancement. The program (including this project) has been awarded a state Fish Transport Permit.

**Mixed-stock Fishery.** The potential for the Port Graham pink salmon hatchery program (including this project) creating or exacerbating a mixed stock fishery program is minimal. The harvest of Port Graham pink salmon are spatially and/or temporally separated from other Kachemak Bay pink salmon stocks as well as other salmon species. There is very little overlap. The same is true with the other salmon species that spawn in the Port Graham area.

**Monitoring and Evaluation.** A portion of the pink salmon reared to 8 grams will be coded wire tagged. The local fisheries and the hatchery egg take will be monitored for marked fish.

**Economic Criteria.** This project, especially long term rearing pink salmon fry to increase adult survival, will negatively impact the hatchery benefit/cost ratio. However, not doing this project would either cause a reduction in the overall subsistence harvest in Port Graham as well as put additional pressure on the wild stocks, and/or extend the hatchery broodstock development phase to the point where operating the hatchery stops making economic sense.

Procedural Criteria. All evaluations (Regional Salmon Planning Team, Coastal Project Certification) of the Port Graham hatchery program (including this project) have been conducted and all necessary permits (hatchery permit, fish transport permit, COE, DNR, CZM) have been obtained. This project has not been evaluated under the NEPA process.

### **C. Cooperating Agencies, Contracts and Other Agency Assistance**

The Port Graham IRA Council will operate this project under a contract with ADF&G. The funds for stream survey air charters will be retained by ADF&G to supplement the normal management surveys of Port Graham.

## **SCHEDULE**

### **A. Measurable Project Tasks for FY 98**

October, 1997	Incubators containing the lots intended for extended rearing and heated water rearing are identified and heat treated to produce a separate otolith mark for each lot.
November	After eye-up eggs from the lot intended to reach 1 gram by late May are put on a heated water regimen.
May, 1998	Heated water rearing lot intended to produce fingerling with average weight of 1 gram are released into zooplankton bloom.
May	Fry receiving standard treatment (incubated and reared in ambient temperature water and held for release into zooplankton bloom) are released into zooplankton bloom.
late June, early July	Lot held for extended rearing in ambient temperature water are released after having reached an average weight of 1 gram.
July 7 to August 31	Monitor pink salmon return to Port Graham.
August 10 to August 25	Capture hatchery broodstock.
August 28 to September 10	Egg take.
April 1999	Annual report on FY 98 work.

### **B. Project Milestones and Endpoints**

The project objective will be successfully met if broodstock development phase is completed on schedule at the end of FY 00.

### **C. Completion Date**

This project will end when the broodstock development phase at the Port Graham hatchery is complete. This is expected to occur by the end of FY 00.

## **PUBLICATIONS AND REPORTS**

Annual reports	Describes project activities for the year, analyzes successes and problems, makes recommendations for improvements due April 1 following fiscal year being reported on.
Final report	Synopsis of each year's activities with analysis of project as a whole. Due April 1 following final year of project.

## **PROFESSIONAL CONFERENCES**

No travel to professional conferences will be paid for out of this project. However, hatchery staff will be attending the Alaska Hatchery Manager's Workshop and the Native American Fish & Wildlife Society meeting and at which they will give a presentation of the work done under this project.

## **COORDINATION AND INTEGRATION OF RESTORATION EFFORT**

If funded, this project will be integrated into the overall pink salmon enhancement program in Port Graham.

## **EXPLANATION OF CHANGES IN CONTINUING PROJECTS**

The production of a lot of fingerling with an average weight of 8 grams has been eliminated because of the high potential for this lot to contract warm water vibrio. In its place heated water will be used to produce a lot of fingerling with an average weight of one gram from release during the mature zooplankton bloom in late May. This lot will test the efficacy of this strategy compared with rearing fry in ambient temperature water until they have achieved an average weight of 1 gram before releasing them.

## **PRINCIPAL INVESTIGATOR**

Ephim Anahonak, Jr., Hatchery Manager  
Port Graham Hatchery  
P. O. Box 5543  
Port Graham, AK 99603  
phone (907) 284-2233  
fax (907) 284-2238

Mr. Anahonak has been hatchery manager of the Port Graham hatchery for the past four years. He has had and will continue to have overall responsibility for the project.

## **OTHER KEY PERSONNEL**

Paul McCollum, hatchery consultant. Mr. McCollum will advise the hatchery staff on the procedures and techniques needed to achieve project objectives.

David Daisy, fish culture consultant. Mr. Daisy will work with the hatchery staff and Mr. McCollum in project design, implementation and reporting.

**FY 98 EXXON VALDEZ TRUST COUNCIL PROJECT BUDGET**

October 1, 1997 - September 30, 1998

Budget Category:	Authorized FFY 1996	Proposed FFY 1997							
Personnel	\$0.0	\$0.0							
Travel	\$0.0	\$0.0							
Contractual	\$69.5	\$71.5							
Commodities	\$0.0	\$0.0							
Equipment	\$0.0	\$0.0							
Subtotal	\$69.5	\$71.5	LONG RANGE FUNDING REQUIREMENTS						
General Administration	\$4.9	\$5.0	Estimated FFY 1999	Estimated FFY 2000	Estimated FFY 2001	Estimated FFY 2001	Estimated FFY 2002	Estimated FFY 2003	
Project Total	\$74.4	\$76.5	\$77.2	\$78.5	\$0.0	\$0.0	\$0.0	\$0.0	
Full-time Equivalents (FTE)		0.0							
Dollar amounts are shown in thousands of dollars.									
Other Resources									
Comments:									

**FY 98**

Project Number: 98225  
 Project Title: Port Graham Pink Dalmon Subsistence Project  
 Agency: AK Dept. of Fish & Game

FORM 3A  
 AGENCY  
 PROJECT  
 DETAIL

Prepared : 4/3/97

1 of 8

Printed: 4/10/97



# FY 98 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1997 - September 30, 1997

Personnel Costs:			GS/Range/ Step	Months Budgeted	Monthly Costs	Overtime	Proposed FFY 1997	
PM	Name	Position Description						
*							0.0	
							0.0	
							0.0	
							0.0	
							0.0	
							0.0	
							0.0	
							0.0	
							0.0	
							0.0	
							0.0	
							0.0	
							0.0	
							0.0	
Subtotal				0.0	0	0		
Those costs associated with program management should be indicated by placement of an *.							<b>Personnel Total</b>	\$0.0

Travel Costs:		Ticket Price	Round Trips	Total Days	Daily Per Diem	Proposed FFY 1997	
PM	Description						
						0.0	
						0.0	
						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 *.						<b>Travel Total</b>	\$0.0

**FY 98**

Project Number: 98225  
 Project Title: Port Graham Pink Salmon Subsistence Project  
 Agency: AK Dept. of Fish & Game

FORM 3B  
 Personnel  
 & Travel  
 DETAIL

**FY 98 EXXON VALDEZ TRUST COUNCIL PROJECT BUDGET**  
 October 1, 1997 - September 30, 1998

<b>Contractual Costs:</b>		Proposed
Description		FFY 1997
Contract with non-trustee agency		71.5
		:
When a non-trustee organization is used, the form 4A is required.		
<b>Contractual Total</b>		<b>\$71.5</b>
<b>Commodities Costs:</b>		Proposed
Description		FFY 1997
<b>Commodities Total</b>		<b>\$0.0</b>

**FY 98**

3 of 8

Project Number: 98225  
 Project Title: Port Graham Pink Salmon Subsistence Project  
 Agency: AK Dept. of Fish & Game

FORM 3B  
 Contractual &  
 Commodities  
 DETAIL

Printed: 4/10/97

October 1, 1997 - September 30, 1998

# FORM 3B Equipment DETAIL

**FY 98 EXXON VALDEZ TRUST COUNCIL PROJECT BUDGET**  
 October 1, 1997 - September 30, 1998

<b>Budget Category:</b>	<b>Authorized FFY 1997</b>	<b>Proposed FFY 1997</b>						
Personnel	\$30.6	\$31.6						
Travel	\$0.0	\$0.0						
Contractual	\$16.5	\$16.5						
Commodities	\$11.4	\$12.4						
Equipment	\$0.0	\$0.0						
Subtotal	\$58.5	\$60.5	LONG RANGE FUNDING REQUIREMENTS					
Indirect	\$11.0	\$11.0	Estimated FFY 1999	Estimated FFY 2000	Estimated FFY 2001	Estimated FFY 2002	Estimated FFY 2003	Estimated FFY 2004
Project Total	\$69.5	\$71.5	\$72.2	\$72.9	\$0.0	\$0.0	\$0.0	\$0.0
Full-time Equivalents (FTE)		1.0						
Dollar amounts are shown in thousands of dollars.								
Other Resources								
Comments:								

**FY 98**

Project Number: 98225  
 Project Title: Port Graham Pink Salmon Subsistence Project  
 Name: Chugach Regional Resources Commission

FORM 4A  
 Non-Trustee  
 DETAIL

October 1, 1997 - September 30, 1998

FY 98

Project Number: 98225  
Project Title: Port Graham Pink Salmon Subsistence Project  
Name: Chugach Regional Resources Commission

FORM 4B  
Personnel  
& Travel  
DETAIL

**FY 98 EXXON VALDEZ TRUST JUNCIL PROJECT BUDGET**  
 October 1, 1997 - September 30, 1998

<b>Contractual Costs:</b>		Proposed
Description		FFY 1997
Freight		1.0
Utilities		1.2
Maintenance & Repair		0.8
Telephone		0.2
Seine boats for broodstock collection 8 days @ \$550/day		4.4
Building Rent - 2months		1.6
Air charter for stream surveys - to ADF&G		2.3
Technical consultants		5.0
<b>Contractual Total</b>		<b>\$16.5</b>
<b>Commodities Costs:</b>		Proposed
Description		FFY 1997
Fish Food		8.5
Skiff fuel/oil		0.3
Plumbing supplies		0.2
Building supplies		0.3
40 x 40 rearing pen nets (2)		3.1
<b>Commodities Total</b>		<b>\$12.4</b>

**FY 98**

7 of 8

Project Number: 98225  
 Project Title: Port Graham Pink Salmon Subsistence Project  
 Name: Chugach Regional Resources Commission

FORM 4B  
 Contractual &  
 Commodities  
 DETAIL

Printed: 4/10/97

October 1, 1997 - September 30 1998

<p><b>FY 98</b></p>	<p>Project Number: 98225          Project Title: Port Graham Pink Salmon Subsistence Project          Name: Chugach Regional Resources Commission</p>	<p>FORM 4B          Equipment          DETAIL</p>
<p>8 of 8</p>		<p>Printed: 4/10/97</p>





## **Project Title; Exhibits on Human Uses of Marine Resources for the Alaska SeaLife Center**

Project Number: 98236

Restoration Category: General Restoration

Proposer: Alaska Native Harbor Seal Commission

Lead Trustee Agency: ADFG  
Cooperating Agencies

Alaska SeaLife Center: Yes

Duration: 1<sup>st</sup> year, 1-year project

Cost FY 98: \$84.6

Cost FY 99: 0

Cost FY 00: 0

Cost FY 01: 0

Cost FY 02: 0

Geographic Area: Spill Area Wide

Injured Resource/Service: All Injured Resources, Subsistence

**RECEIVED**  
APR 11 1997

**EXXON VALDEZ OIL SPILL  
TRUSTEE COUNCIL**

### **ABSTRACT**

Alaska Native residents of the oil spill impact area have expressed the opinion that it is important that information on their harvest and use of marine resources, including marine mammals, seabirds, invertebrates and fish, be incorporated into the exhibits presented at the Alaska SeaLife Center. This project proposes to produce educational exhibits on the human uses of the various marine animals on display at the Sea Life Center.

### **INTRODUCTION**

This project proposes to produce educational exhibits that illustrate the use of natural resources by indigenous residents of Alaska coastal regions, especially the *Exxon Valdez* oil spill impact area. These exhibits could include short videos, artifacts, hunting and processing implements, crafts, photographs and explanatory text.

The project is being proposed in response to concerns expressed by Alaska Native residents of the oil spill impact area, as well as other coastal areas in the state, over the possible impact the exhibits and information presented at the Alaska SeaLife Center in Seward could have on their subsistence practices.

The subsistence service itself was injured by the oil spill, and there has been a substantial amount of time and funding devoted by the *Exxon Valdez* Oil Spill Trustee Council to restore this service. Alaska Native residents of the oil spill impact area have expressed the opinion that it is important that information on their harvest and use of marine species be incorporated into the exhibits presented at the SeaLife Center to prevent further injury to subsistence and to promote the recovery of subsistence.

## **NEED FOR THE PROJECT**

### **A. Statement of Problem**

Concern has arisen among the Alaska Native residents of the oil spill impact area, as well as other coastal areas in the state, over the possible impact the exhibits and information presented in the public education portions of the Alaska SeaLife Center in Seward could have on public perceptions of marine resources and Alaska Native lifestyles. Residents fear that if these exhibits are not presented in the proper context, there will be increased pressure on Alaska Natives to curtail or cease their harvests and uses of these resources. This would have a further deleterious effect on the subsistence service, which was injured by the oil spill and its aftermath.

This project would mitigate these possible negative impacts to subsistence by educating the public about Alaska Native lifestyles and subsistence uses of marine resources. This will also be a positive step towards restoration of subsistence. This work would also support cultural values, including sharing the sharing of information, and it promote involvement in the restoration process.

### **B. Rationale/Link to Restoration**

Alaska Native residents of the oil spill impact area have expressed the opinion that it is important that information on their harvest and use of marine resources be incorporated into the exhibits presented at the SeaLife Center. This project proposes mitigate the potential for additional negative impacts to the subsistence service. It will do this by educating the public about subsistence uses of marine resources and Alaska Native lifestyles in a manner considered appropriate by Alaska Natives. This project will provide members of the Alaska Native community the opportunity they have requested for involvement in the development of interpretive exhibits at the Alaska SeaLife Center. It will also provide for the incorporation of traditional and local ecological knowledge concerning marine resources in the exhibits.

### **C. Location**

The Alaska SeaLife Center, where the proposed exhibits would be displayed is located in Seward, Alaska. The proposed exhibits will represent traditional, cultural, and subsistence

practices from throughout the coastal areas of Alaska, and especially the oil spill impact area, which includes Prince William Sound, Kodiak, the lower Kenai Peninsula and the Alaska Peninsula.

## **COMMUNITY INVOLVEMENT AND TRADITIONAL ECOLOGICAL KNOWLEDGE**

This project will provide the involvement requested by the Alaska Native community in the development of interpretive exhibits at the Alaska SeaLife Center. It will also provide the opportunity for traditional and local ecological knowledge to be incorporated into the exhibits.

The guidance of the Spill Area Wide coordinator and the community facilitators will be sought in the design of the interpretative exhibits.

## **PROJECT DESIGN**

### **A. Objectives**

1. Respond to concerns expressed by members of the Alaska Native community over the content of exhibits at the Alaska SeaLife Center.
2. Provide an opportunity for Alaska Native involvement in the development of interpretive exhibits at the Alaska SeaLife Center.
3. Mitigate potential negative impacts to the subsistence service from information presented at the Alaska SeaLife Center.
4. Integrate traditional and local ecological knowledge on marine resources into exhibits at the SeaLife Center.
5. Educate the visiting public about Alaska Native lifestyles and subsistence uses of marine resources in a manner considered appropriate by the Alaska Native community.

### **B. Methods**

ADG&G will contract with the Alaska Native Harbor Seal Commission to organize a five member committee to oversee the content and design of the proposed exhibits. The committee will be made up of representatives from the various coastal regions of the state of Alaska, including a representatives from Southeast Alaska, Southcentral Alaska, the Kodiak region, the Bristol Bay region, and the Nome region.

The oversight committee would have two meetings in Anchorage, the first to review the proposals received for the video and design contracts, and the second to review and comment on the work of the contractors prior to construction and installation of the exhibits.

In addition, the principal investigators will consult with the Alaska SeaLife Center, regional Alaska Native organizations (including the Alaska Native Harbor Seal Commission, the Steller Sea Lion Commission and the Alaska Sea Otter Commission), coastal village councils, the Spill Area Wide Community Involvement Coordinator, and the community facilitators on the content and design of the proposed interpretive exhibits.

Contracts will be let for the design and construction of the exhibits, and for production and post-production video work. Most of the video segments needed for the exhibits can be culled from existing videos (for example, excerpts from "Alutiiq Pride: A Story of Subsistence" could be used for the harbor seal exhibit). However, it may be necessary for some original footage to be shot to cover gaps in the available video footage.

### **C. Cooperating Agencies, Contracts and Other Agency Assistance**

Contracts will be let for the design and construction of the exhibits, and for production and post-production video work.

## **SCHEDULE**

### **A. Measurable Project Tasks for FY 98 (October 1, 1997-September 30, 1998)**

October 1, 1997	Request for proposals issued for exhibit design and construction Request for proposals issued for production and post-production of videos
November 1, 1997	Contract with Alaska Native Harbor Seal Commission in place Members of oversight committee selected and in place
November 15, 1997	Oversight committee meets to review proposals received for exhibit design and construction, and production and post-production of videos
December 1, 1997	Contract awarded for exhibit design and construction Contract awarded for production and post-production of videos
January 15, 1997	Oversight committee meets with contractors to review and comment on exhibit design plans and video story boards
March 1, 1998	Video production and post-production complete
April 1, 1998	Exhibit design complete
May 1, 1998	Exhibit construction and installation complete
May 1998	Alaska SeaLife Center opens
April 15, 1999	Final report describing consultative process submitted
On-going project task	Consultation with Alaska SeaLife Center, regional Alaska Native organizations, coastal Alaska village councils, Spill Area-wide Community Involvement Coordinator and community facilitators.

### **B. Project Milestones and Endpoints**

1. The response to concerns expressed by members of the Alaska Native community over the content of exhibits at the Alaska SeaLife Center will be achieved with the on-going consultations with representatives of that community, these consultations will be complete with the opening of the SeaLife Center (May 1998).
2. The opportunity for Alaska Native involvement in the development of interpretive exhibits at the Alaska SeaLife Center will be met when the ideas and concerns expressed in those consultations are translated into interpretative exhibits (by April 1, 1998).
3. The mitigation of potential negative impacts to the subsistence service of information presented at the Alaska SeaLife Center will be achieved with the installation of the interpretative exhibits in May 1998.
4. The integration of traditional and local ecological knowledge on marine mammals into exhibits at the SeaLife Center will be achieved with the completion of the videos and exhibit design (April 1, 1998).
5. Education of the visiting public about Alaska Native lifestyles and subsistence uses of marine resources in a manner considered appropriate by the Alaska Native community will, in one sense, be achieved with the opening of the Alaska SeaLife Center (May 1998), in another sense this education will continue as long as the SeaLife Center continues to admit visitors and as long as the interpretative exhibits remain in place.

### **C. Completion Date**

The project will be complete in federal fiscal year 1998, the objectives of the project will continue to be achieved as long as the interpretative exhibits remain on display to the visiting public.

### **PUBLICATIONS AND REPORTS**

No publications are anticipated as a result of this project. A final report describing the consultation process undertaken in the project will be submitted to the Trustee Council on April 15, 1999.

### **PROFESSIONAL CONFERENCES**

No attendance at professional conferences is anticipated as part of this project.

### **COORDINATION AND INTEGRATION OF RESTORATION EFFORT**

Through the consultation process, this project will be coordinated with the Alaska SeaLife Center, the Community Involvement Project and the Traditional Ecological Knowledge Project.

## PROPOSED PRINCIPAL INVESTIGATORS

Monica Riedel  
Alaska Native Harbor Seal Commission  
P.O. Box 2229  
Cordova, Alaska 99574  
Phone number: 907-424-5882  
Fax number: 907-424-5883

Rita A. Miraglia  
Alaska Department of Fish and Game  
Division of Subsistence  
333 Raspberry Road  
Anchorage, Alaska 99518  
Phone number: 907-267-2358  
Fax number: 907-267-2450  
E-mail address: RitaM@fishgame.state.ak.us

## PRINCIPAL INVESTIGATORS

Monica Riedel Ms Riedel, an Alaska Native resident of Cordova, is the Executive Director of the Alaska Native Harbor Seal Commission, and the Vice-Chair of the Indigenous Peoples Conference on Marine Mammals. She has extensive experience in marine mammal issues through her work with the Alaska Sea Otter Commission. She is responsible for the ANHSC activities under 98244, including identifying and subcontracting with local village technicians, developing subcontracts, and participating in workshops and community meetings.

Rita Miraglia: Ms Miraglia has served as the oil spill coordinator for the Division of Subsistence since 1990. As such, she has organized and participated in the subsistence resource collection and testing programs of 1990 and 1991, and participated in the community based subsistence restoration planning process begun in 1994. She has served as the Division's primary liaison with the Oil Spill Health Task Force, and communicated restoration study findings to the communities in the oil spill area through community meetings and newsletters. Ms Miraglia has a Masters degree in Anthropology from the State University of New York. Before coming to the Division, she worked for Chugach Alaska Corporation. As a member of CAC's Oil Spill Response Team, Ms Miraglia sat on the Interagency Shoreline Clean-up Committee in Valdez in 1989, and the Cultural Technical Advisory Group in 1990, working to ensure that the concerns of the predominantly Alaska Native communities and native regional organizations were considered in the oil spill response.



UNIVERSITY OF ALASKA FAIRBANKS  
Fairbanks, Alaska 99775-1080

April 4, 1997

EVOS Trustee Council  
Anchorage, Alaska

To whom it concerns:

I have had the opportunity review the proposal entitled *Exhibits on human use of marine resources for the Alaska SeaLife Center* and I wanted to urge your careful consideration of this package. I have worked with the Native community on EVOS funded projects looking at the health of harbor seals in Alaska and have spent a good deal of time in discussions on the role of marine mammals in their culture. I agree with them that the role of marine mammals in their culture is unique in the United States and that this needs to be emphasized to the public whenever possible. The Alaska SeaLife Center will be a focal point for visitors coming to Alaska where many different issues of marine life will be presented. It makes sense to me that some sort of acknowledgment to the role of marine mammals in the Native community should be available at the ASLC. This proposal represents such an acknowledgment and could form the basis for a very important display.

I also understand that such displays will need the approval of the Education division, Executive director and possibly the Board of Directors of the ASLC, but I feel that this proposal is a critical step in that direction and that it should be seriously considered.

Sincerely,

A handwritten signature in black ink, appearing to read "Michael Castellini", written over a horizontal line.

Dr. Michael Castellini

# 1998 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1997 - September 30, 1998

Budget Category:	Authorized FY 1997	Proposed FY 1998						
Personnel		\$8.0						
Travel		\$1.2						
Contractual		\$69.3						
Commodities		\$0.0						
Equipment		\$0.0						
Subtotal	\$0.0	\$78.5	LONG RANGE FUNDING REQUIREMENTS					
General Administration		\$6.1		Estimated FY 1999	Estimated FY 2000	Estimated FY 2001	Estimated FY 2002	
Project Total	\$0.0	\$84.6		\$0.0	\$0.0	\$0.0	\$0.0	
Full-time Equivalents (FTE)		0.1						
Dollar amounts are shown in thousands of dollars.								
Other Resources								
Comments:								

1998

Project Number: 98236  
 Project Title: Exhibits on Human Uses of Marine Resources for the  
 Alaska SeaLife Center  
 Agency: Alaska Department of Fish and Game

FORM 3A  
 TRUSTEE  
 AGENCY  
 SUMMARY



# 1998 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1997 - September 30, 1998

Personnel Costs:		GS/Range/ Step	Months Budgeted	Monthly Costs	Overtime	Proposed FY 1998
Name	Position Description					
Rita Miraglia	Subsistence Resource Specialist III	18C	1.5	5.3	0.0	8.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
Subtotal			1.5	5.3	0.0	
Personnel Total						\$8.0
Travel Costs:		Ticket Price	Round Trips	Total Days	Daily Per Diem	Proposed FY 1998
Description						
Anchorage-Seward		0.1	4	8	0.1	1.2
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
Travel Total						\$1.2

1998

Project Number: 98236  
 Project Title: Exhibits on Human Uses of Marine Resources for  
 the Alaska SeaLife Center  
 Agency: Alaska Department of Fish and Game

FORM 3B  
 Personnel  
 & Travel  
 DETAIL

Prepared: 2 of 8

4/10/97

# 1998 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1997 - September 30, 1998

<b>Contractual Costs:</b>		Proposed
Description		FY 1998
4A Linkage	Contract with Alaska Native Harbor Seal Commission to organize oversight committee	19.3
	Contract with museum exhibit designer (includes materials to construct exhibits	40.0
	Contract for production/post-production video	10.0
When a non-trustee organization is used, the form 4A is required.		
<b>Contractual Total</b>		<b>\$69.3</b>
<b>Commodities Costs:</b>		Proposed
Description		FY 1998
<b>Commodities Total</b>		<b>\$0.0</b>

**1998**

Project Number: 98236  
 Project Title: Exhibits on Human Uses of Marine Resources for the  
 Alaska SeaLife Center  
 Agency: Alaska Department of Fish and Game

FORM 3B  
 Contractual  
 & Commodi  
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Prepared:

3 of 8

4/10/97

**1998 EXXON VALDEZ TRUSTEE INCIL PROJECT BUDGET**  
October 1, 1997 - September 30, 1998

[illegible]

## 1998

Project Number: 98236  
Project Title: Exhibits on Human Uses of Marine Resources for  
the Alaska SeaLife Center.  
Agency: Alaska Department of Fish and Game

FORM 3B  
Equipment  
DETAIL

# 1998 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1997 - September 30, 1998

Budget Category:	Authorized FY 1997	Proposed FY 1998						
Personnel		\$8.0						
Travel		\$8.8						
Contractual		\$0.0						
Commodities		\$0.0						
Equipment		\$0.0	LONG RANGE FUNDING REQUIREMENTS					
Subtotal	\$0.0	\$16.8		Estimated FY 1999	Estimated FY 2000	Estimated FY 2001	Estimated FY 2002	
Indirect		\$2.5						
Project Total	\$0.0	\$19.3						
Full-time Equivalents (FTE)		2.0						
Dollar amounts are shown in thousands of dollars.								
Other Resources								
Comments:								

**1998**

Project Number: 98236  
 Project Title: Exhibits on Human Uses of Marine Resources for the  
 Alaska SeaLife Center  
 Name: Alaska Native Harbor Seal Commission

FORM 4A  
 Non-Trustee  
 SUMMARY

**1998 EXXON VALDEZ TRUSTEE INCIL PROJECT BUDGET**  
**October 1, 1997 - September 30, 1998**

<b>Personnel Costs:</b>			Months Budgeted	Monthly Costs	Overtime	Proposed FY 1998
Name	Position Description					
Monica Riedel	Executive Director		2.0	4.0	0.0	8.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
Subtotal			2.0	4.0	0.0	6.0
<b>Personnel Total</b>						<b>\$8.0</b>
<b>Travel Costs:</b>		Ticket Price	Round Trips	Total Days	Daily Per Diem	Proposed FY 1998
Description						
Anchorage-Dillingham		0.5	2	2	0.1	1.2
Anchorage-Nome		0.6	2	2	0.1	1.4
Anchorage-Kodiak		0.5	2	2	0.1	1.2
Anchorage-Cordova		0.3	6	8	0.1	2.6
Anchorage-Juneau		0.5	2	2	0.1	1.2
Anchorage-Seward		0.1	4	8	0.1	1.2
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
<b>Travel Total</b>						<b>\$8.8</b>

## 1998

Project Number: 98236  
Project Title: Exhibits on Human Uses of Marine Resources for  
the Alaska SeaLife Center  
Name: Alaska Native Harbor Seal Commission

FORM 4B  
Personnel  
& Travel  
DETAIL

Prepared: 6 of 8

4/10/97

# 1998 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1997 - September 30, 1998

<b>Contractual Costs:</b>		<b>Proposed FY 1998</b>
<b>Description</b>		
<b>Contractual Total</b>		<b>\$0.0</b>
<b>Commodities Costs:</b>		<b>Proposed FY 1998</b>
<b>Description</b>		
<b>Commodities Total</b>		<b>\$0.0</b>

**1998**

Project Number: 98236  
 Project Title: Exhibits on Human Uses of Marine Resources for  
 the Alaska SeaLife Center  
 Name: Alaska Native Harbor Seal Commission

FORM 4b  
 Contractual  
 & Commodi  
 ties

Prepared:

7 of 8

4/10/97

**1998 EXXON VALDEZ TRUSTEE NCIL PROJECT BUDGET**  
**October 1, 1997 - September 30, 1998**

New Equipment Purchases:		Number of Units	Unit Price	Proposed FY 1998
Description				
				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 a <b>New</b>				0.0
<b>Existing Equipment Usage:</b>			Number of Units	
Description				

## 1998

Project Number: 98236  
Project Title: Exhibits on Human Uses of Marine Resources for  
the Alaska SeaLife Center  
Name: Alaska Native Harbor Seal Commission

FORM 4B  
Equipment  
DETAIL

Prepared: 8 of 8

4/10/97





Salmon carcasses and juvenile Chinook salmon production  
in the Kenai River Ecosystem  
FY 98 Detailed Project Description

RECEIVED  
APR 11 1997

Project Number: 98239

Restoration Category: Monitoring/Restoration

Proposer: Alaska Department of Fish and Game

Lead Trustee Agency: Alaska Department of Fish and Game  
Cooperating Agencies: U.S.F.W.S, N.B.S., Department of Interior

Duration: Year 1 of 2

Cost FY 98: \$166,600  
Cost FY 99: \$100,000  
Cost FY 00: \$0

EXXON VALDEZ OIL SPILL  
TRUSTEE COUNCIL

Geographic Area: Kenai Peninsula

Injured Resource/Service: This project is intended to evaluate the impacts of the large 1989 escapements on juvenile chinook salmon production rates by studying nutrient trophic dynamics in the Kenai River and selective tributaries.

## ABSTRACT

This proposal provides for an investigation as to the role sockeye salmon carcasses play in primary and secondary production within the mainstem Kenai River. Using recently developed methodologies, we propose to expand our investigations into the potential symbiotic role sockeye salmon escapements have on nutrients and secondary productivity of the mainstem Kenai River. Investigations to date have involved only the direct effect of adult escapements and their progeny have had on the Kenai glacial lake ecosystems and their interaction with the zooplankton community and interactions among adjacent year classes of both sockeye salmon and the dominant zooplankton species. The development of a holistic ecosystem approach to restoration of this system requires examination of the role salmon carcasses play in freshwater life history of other important commercial species. Chinook salmon production may be positively influenced by nutrient additions to the relatively oligotrophic waters of the Kenai River. Although there are four anadromous salmon runs into the Kenai, sockeye salmon are the largest in terms of adult biomass. This study will focus on determining if measurable benefits to chinook salmon growth rates can be attributed to salmon carcasses in general, and more specifically, sockeye salmon effects. **The fundamental question to be addressed the first year by this program is to determine if there is a sufficient marine derived nutrient component that can be measured in a large glacial river.** This aspect of the ecology of the Kenai River has had much interest by

members of the public, members of the Board of Fisheries, the U.S. Fish and Wildlife Service and ADF&G staff. The method of restoration of this system will most likely be confined to regulation of harvest rates of the fisheries. An important feature of these studies is to ascertain if there are significant benefits to chinook salmon juveniles with increased escapements. In addition, this program has the side benefit of providing insight as to historical significance of the dependence of the current chinook salmon fishery on the historically recent increase in sockeye salmon runs to the Kenai.

## INTRODUCTION

The Trustee Council has recently invested millions of dollars in the restoration of streambanks on the Kenai River. This habitat is dominantly used by juvenile chinook (king) salmon during the summer freshwater growth period. This investigation will examine if the chinook salmon population in the Kenai River receives much of its productivity from salmon carcasses. In addition to the cover provided for rearing juvenile chinook salmon from bankside habitat, the amount of food available may well be determined by the nutrient status of the river. Although other species, such as coho salmon, rainbow trout, Dolly Varden, stickleback, and sculpin are important rearing species in the Kenai drainage and undoubtedly have life history elements influenced by salmon carcasses, we focus on chinook salmon for several reasons. Their rearing life history is completely within carcass influenced portions of the Kenai River and their diet is based on secondary production rather than direct consumption of juvenile salmon, salmon eggs or fish carcasses. Adult returns of Kenai River chinook salmon are most likely dependent upon freshwater survival during their first year of emergent life in the river. First year growth and the resultant overwintering success most likely determine the status of Kenai chinook salmon runs. Other species of commercial importance have extended freshwater life histories and studies of their population dynamics are compounded by their spending much of their life history in habitats outside of the Kenai mainstem. These characteristics would make isolating the role of fish carcasses on productivity much more problematic. In addition, costs of the investigations would go up considerably as the study plan would require near replication for each species and the problem of most interest with resident species is most likely associated with growth rates

Although chinook salmon were not a species directly damaged by the oil spill, their life history may be intrinsically tied to sockeye salmon and chinook salmon are clearly associated with sports, subsistence, and commercial fisheries in upper Cook Inlet that were adversely affected by EVOS. Studies on the Kenai River since 1990 have addressed the large escapements of sockeye salmon into the Kenai River following commercial fisheries closures (Schmidt and Tarbox, 1993; 1995; 1996a; 1996b). These studies have focused on the population dynamics and trophic interactions of sockeye salmon juveniles rearing in the major glacial lake on the Kenai River. The limnological analyses (Schmidt and Tarbox 1996b) have established the biological mechanism for much of the observed variation in sockeye salmon run returns. In addition, investigations focused on the all trophic levels within the lake, including the influence of sockeye salmon carcasses on the productivity of these lakes. One of the techniques used in these investigations is the examination of carbon and nitrogen isotopes that are much more abundant in the marine food chain and concentrate in adult salmon carcasses (Kline et al. 1990; 1994). Smolt samples collected from the glacial lakes indicated relatively low levels of these isotopes in

sockeye salmon smolts that have reared in these lakes. On a comparative basis, these lakes had very low isotope levels when compared with other systems, such as Lake Illiamna, Red Lake, or Karluk Lake. This is explainable by two factors. Adult sockeye salmon densities per unit area of these lakes is quite low when compared with the major clear water sockeye salmon systems (Personal Comm, B. Finney, U. of Alaska, Fairbanks). In addition, much of the spawning population of Skilak Lake, the dominant producer, is within the outlet. This most likely results in the nutrients being dispersed in the Kenai River below Skilak Lake. The development of a restoration strategy based on regulation of escapements into the Kenai River is incomplete without a broader based view as to the impact of these carcasses on the productivity of the mainstem Kenai River with particular focus on the chinook salmon population of the Kenai. This type of study was identified by a review team when examining the Kenai River salmon issues prior to the 1996 spring Board of Fisheries meetings and is supported by the Kenai Peninsula office of USFWS (See attachment 1). This information will provide a quantitative assessment as to the broader ecological role sockeye salmon carcasses play in the Kenai River ecosystem and will provide a better perspective as to the impacts of large escapements on the overall production in the Kenai River and aide in the determining if there are tradeoffs in establishment of escapement.

## **NEED FOR THE PROJECT**

### **A. Statement of Problem**

The previous investigations into the role of large escapements of sockeye salmon on the Kenai River have resulted in an improved understanding of the density dependent responses of sockeye salmon and their ecological consequences to the large glacial lakes of the Kenai River. Public and agency questions have arisen concerning the breadth of these studies in that resident and other anadromous fish species within the Kenai River may have indirectly benefited from the larger sockeye salmon escapements. Although all species of fish within the Kenai River undoubtedly have some interaction with sockeye salmon carcasses, the magnitude of this impact is highly speculative. This study proposes to focus on juvenile chinook salmon production in the Kenai River, and will determine if the abundance of salmon carcasses can be related to their productivity. These data will be used in the development of escapement goals in the Kenai system and will provide agencies with added information to assess if bottom up trophic level benefits to the Kenai River provide for a measurable offset to the losses of yield associated with overescapement of sockeye salmon. In addition, the maintenance of large sustainable runs of sockeye salmon to the Kenai through proper management of sockeye stocks, may be significantly related to the chinook salmon returns to the Kenai. This will provide the public with information as to the degree of dependence management of one of these stocks is on the successful propagation of the other, and will provide management agencies with insight as to the impact harvest management population restoration will have on other important economic salmon species.

### **B. Rationale/Link to Restoration**

The proposed study will examine other roles sockeye salmon escapements have on in river ecological processes as opposed to the concentration on the lake ecosystems. In the development of restoration of sockeye salmon through escapement goal development, a broader ecosystem perspective on escapements that have the potential to affect populations of other species is desirable. Although many bird, mammal, and fish species may have an interaction with sockeye salmon carcasses, chinook salmon are highly dependent upon the mainstem rearing environment and have a major economic value to the economy of the Kenai Peninsula. The relatively recent development of stable isotope techniques have provided a method of quantifying the trophic transfer of carcass based nutrients in lakes and rivers (Kline et al. 1990; 1994; Rand et al. 1992; Bilby et al. 1996; Piorkowski 1995). In addition, the Alaska Department of Fish and Game has conducted studies on the distribution and migration of juvenile chinook salmon within the Kenai River. The current hypothesis on the factors limiting chinook salmon production in the Kenai River suggests that the smolt production is dependent upon juvenile chinook (rearing within the river) obtaining sufficient size to effectively migrate to suitable overwintering habitat within the system (Personal Comm. Terry Bendock, ADF&G, Soldotna). This study will examine the  $\delta^{15}\text{N}$  and  $\delta^{13}\text{C}$  of juvenile chinook salmon rearing within different reaches and tributaries of the Kenai River which are affected by different relative abundance of adult salmon carcasses. By comparison of population growth rates prior to the introduction of salmon carcasses and after their introduction, coupled with the temporal and spatial comparisons of stable isotope ratios, we will draw inferences as to the relative importance salmon carcasses contribute to the growth of juvenile chinook salmon in these different reaches of the Kenai. With coho salmon on small stream in the Washington area, Bilby et al. (1996) found major effects of salmon carcass nutrients on growth rates. Although the morphology and volume of the Kenai River is substantially different, these investigations suggest that carcass effects on the food chain may be significant in these systems.

### **C. Location**

These investigations will be confined to the Kenai River drainage and their tributaries. Benefactors of this project will be the citizens of the Kenai Peninsula and other members of the public throughout the railbelt area who utilize the fisheries resources of this system.

## **COMMUNITY INVOLVEMENT**

Residents of the Kenai Peninsula Borough are an important part of the Trustee Council funded Kenai River restoration projects. Besides working on the projects in direct employment as ADF&G Fish and Wildlife biologist and technicians the people of the Peninsula are kept well informed about these projects. Major media outlets in Anchorage and Kenai cover the issues impacting the Kenai River, including the EVOS funded projects. In addition, local ADF&G project biologists have made presentations on restoration efforts to local governments, in local schools, and to community groups. Further, detailed discussions and program suggestions have resulted from the involvement of the Upper Cook Inlet Regional Planning Team. This team is composed of members from the Cook Inlet Regional Aquaculture Association and ADF&G. The team has held numerous meetings with diverse public participation to discuss the results to date

of the EVOS Kenai River projects. The proposed project will be reviewed and subject to feedback on goals and objectives throughout the community.

## **PROJECT DESIGN**

### **A. Objectives**

This study proposal examines the following hypotheses:

1. The growth rate of juvenile chinook salmon will measurably change with the seasonal incremental increase in nutrients from salmon carcasses.
2. Juvenile chinook salmon tissue concentrations of stable isotopes reflect the relative abundance of salmon carcasses among the reaches of the Kenai River and the Kenai River tributaries in which they have reared.
3. Juvenile chinook salmon diets and the trophic status of their diet will provide information sufficient to define the proportion of their growth attributable to salmon carcass derived nutrients.
4. ADF&G escapement data and observations of the relative distribution of salmon spawners within the Kenai River ecosystem will approximate the relative contribution that escapements of the various anadromous species contribute to the productivity of juvenile chinook salmon.

To test these concepts, we will initiate a data collection effort with the following objectives:

- A. Measure the population growth rate of juvenile chinook salmon by obtaining systematic samples on bi-weekly intervals through the growing season after July 1.
- B. Obtain and process samples for marine derived nutrients (stable isotopes) of juvenile chinook salmon from five reaches of the Kenai River and its tributaries with variable adult salmon spawning densities before and after carcasses are assimilated into the system.
- C. Obtain and process samples for stable isotope analysis of various trophic levels of the Kenai River food web prior to and after assimilation of carcasses, paralleling the juvenile chinook samples from Objective B. Food habits will be evaluated by examination of stomach contents.
- D. Obtain from the 1997 and previous historical data bases of ADF&G the relative spawner distribution of the anadromous species present in the Kenai River.

These objectives are designed to provide the needed information for subsequent analysis of the relative importance salmon carcasses have in the trophic status of the mainstem Kenai River. The development of a restoration strategy involving escapement goal adjustment and improved harvest regulation that were funded previously by the Trustees has left some important

information voids. Restoration of sockeye salmon populations through harvest management controls and by the establishment of escapement goals may be highly successful in maintaining strong runs to the Kenai River. However, the net effect of these activities on the potential productivity of other species is the subject of much speculation. In addition, large cyclic runs of sockeye salmon that establish as a result of escapements beyond the carrying capacity of the natal lakes may coincidentally adversely affect other species during the low ebb of their cycles; particularly if we see a density independent change in marine survival. For example, escapements of sockeye salmon during the 1970's in the Kenai River were far below the targeted escapement of the past decade. A successful run of sockeye salmon may be related to the successful production of other species. Alternatively, the volume of the Kenai River and its glacial nature, may render carcass nutrients as a minor component in the overall trophic structure of the river. This is consistent with the low  $\delta^{15}\text{N}$  levels observed in sockeye smolts from Skilak Lake and the initial values from sediment core analysis (Personal Comm. B. Finney, U. of A. Fairbanks, AK.). These analyses should prove useful information on the ongoing discussions concerning biological escapement goals of the Kenai River and debates among the interdependence of sockeye and chinook salmon. We will also estimate the relative contribution of carcasses to the nutrient composition of other elements of the Kenai River food web. This will have implications as to the importance of carcasses on the productivity of other predators not investigated, such as resident fish, birds, and mammals.

## **B. Methods**

The study design proposed here will be refined during the fall and winter of 1997, prior to initiating field investigations.

The study design will be confined to the Kenai River drainage. Five reaches of the Kenai River will be selected during the summer and fall of 1997 to provide the following:

1. One reach of a tributary with minimal upstream spawning salmon but with sufficient rearing juvenile chinook to provide adequate samples for stable isotope and growth analysis without effecting the population
2. One reach of a barriered stream (no anadromous influence) with rearing resident fish (i.e. rainbow trout) that can be used as a control for each trophic level sampled for stable isotopes only.
3. Two reaches of the lower Kenai River mainstem, one immediately below the large sockeye salmon spawning aggregations near Skilak Lake outlet and one below the Soldotna Bridge but above tide water.
4. One reach between the Kenai and Skilak Lakes but above the confluence of the Russian River.

Chinook salmon juveniles will migrate extensively within a reach in response to initial emergence location, subsequent growth, and changes in habitat (Bendock 1989). Therefore, we will not attempt to relate fish characteristics to local micro-habitat, as much of their growth may be a function of previous locales. Samples will be collected from a minimum of five locations

within the river reach. Location of samples will be retained in the analysis and will be used to provide an estimate of sample variance for a reach of river. Five fish per sampling location will be analyzed for  $\delta^{15}\text{N}$  for a time period immediately before spawning and in the fall prior to major migratory movements of juvenile chinook to overwintering areas. All fish collected will be measured for AWL, and a representative sub-sample will be taken for stomach content analysis. Knowledge of the relative distribution of the various salmon carcasses will be used to infer the contribution of the different species to overall productivity of the various reaches.

At each station within each reach a sample of major trophic categories (following the classification of Bisson et al. (1996) and Piorkowski (1995)) will be obtained for stable isotope analysis. The invertebrate species selected will be based on initial stomach analysis of diet. In addition, a sample of organic matter from epithetic organic matter from the stream bed will also be included. Invertebrates will also be collected and preserved for further analysis. . All samples will be processed for stable isotopes following the methods of Kline et al. 1994 by the University of Alaska, Fairbanks under the directive of Dr. Bruce Finney.

Each station will be sampled every 3 weeks from July 1 through October to determine nutrient status following the methods of Koenings et al. (1987) and Litchfield and Kyle (1991). In addition, juvenile chinook salmon will be sampled for AWL at each site (excluding the non-anadromous site) over the summer to establish population growth rates.

If sufficient marine nitrogen is present we will examine the methodology proposed by Bilby (Personal Comm., North Pacific International Chapter Meeting of the American Fisheries Society, March 1997) for establishing escapement goals. This methodology requires development of asymptotic curves of marine nitrogen incorporation into salmon fry as a function of relative carcass contribution to the system.

### **C. Cooperating Agencies, Contracts and Other Agency Assistance**

Administrative support is provided by the Administrative Division, Habitat Division, Sport Fish Division and Commercial Fisheries Management and Development Division staff of the Alaska Department of Fish and Game. The project leaders and their assistants have not been funded by this project and are supported with general funds from the State of Alaska. As we anticipate major budget cuts in FY97 to the limnology program, we have provided for a limited funding of these positions to prepare a peer review publication. Most laboratory analyses are conducted by the limnology laboratory in Soldotna. These studies are integrated with ongoing studies by the Commercial Fisheries Management and Development Division on the Kenai Peninsula. These studies have different objectives, i.e. to manage, enhance, and rehabilitate common property salmon fisheries, but use the same techniques and data collection methods. Consequently, the EVOS investigations have been integrated into the normal operations of these Divisions for efficiency in completing the objectives of these studies and the general mission of these agencies. The stable isotope analysis will be conducted at the University of Alaska, Fairbanks, under the direction of Dr. Bruce Finney. An RSA will be used as the contracting mechanism with ADF&G. This contract will include obtaining study design and project review assistance from Dr. Finney. In addition, these investigations will be coordinated with studies with other

objectives to insure common sampling sites and efficient use of samples will occur. Possible additional studies include water quality and invertebrate studies contracted by Trout Unlimited,

The proposed study provides for data collection and field sampling programs. As such no environmental effect of these programs occurs beyond that of traditional fisheries management data collection activities, and is within existing collecting permits or Federal special use permits issued to the Department of Fish and Game for scientific data collection activities. New programs on the Kenai National Wildlife Refuge are updated through permit amendments as needed. No other permits or other coordination activities are involved.

## **SCHEDULE**

### **A. Measurable Project Tasks for FFY 98-99**

July 1-November 30:	Measure the population growth rate of juvenile chinook salmon by obtaining systematic samples on bi-weekly intervals through the growing season. Completion date would be early winter of 1998.
August 1-April 15:	Obtain and process samples for marine-derived nutrients (stable isotopes) of juvenile chinook salmon from five reaches of the Kenai River and its tributaries with variable adult salmon spawning densities before and after carcasses are assimilated into the system.
May 1-April 15:	Obtain and process samples for stable isotope analysis of various trophic levels of the Kenai River food web prior to and after assimilation of carcasses, paralleling the juvenile chinook samples from objective B. Food habits will be evaluated by examination of stomach contents.
May 1-December 15:	Obtain from the 1998 and previous historical data bases of ADF&G, the relative spawner distribution of the anadromous species present in the Kenai River.

### **B. Project Milestones and Endpoints**

April 15, 1998:	Finalize study design and initiate field work:
April 15, 1999:	Complete draft final report
June 15, 1999:	Submit peer manuscript and final report

### **C. Completion Date**

The project will be completed on September 30, 1999.

## **PUBLICATIONS AND REPORTS**

An annual status report detailing project results for the Kenai River carcass nutrient study will be prepared for peer review on April 15, 1998. This report will entail the final study design that will



be implemented during the field season of 1998. A final separate report, detailing the results of these studies will be issued on April 15, 1999.

## **PROFESSIONAL CONFERENCES**

During the initial year of this contract, no conferences pertinent to this aspect of the study are planned. We anticipate presentation of these findings at scientific meetings in the second year of the investigations.

## **NORMAL AGENCY MANAGEMENT**

The Alaska Department of Fish and Game has ongoing sportfish and commercial fisheries research operations on the Kenai River. In addition, the CFMD Division has ongoing data collection activities from Hidden Lake relating to the limnology of this system. These data are integrated into statewide or regional data bases that are used to directly assess the impacts of the oil spill. In addition, the area research and management biologists for the Division of Commercial Fisheries Management and Development and numerous administrative and support staff are supported by general funds provided by the Alaska legislature. To date, most of the data analysis and reporting for the sockeye salmon over-escapement project has been provided for from contributions of the State of Alaska from these general funds. Total funding for these programs exceeds \$1 million. The proposed investigation is not a normal part of ADF&G's activities.

## **COORDINATION AND INTEGRATION OF RESTORATION EFFORT**

The investigations of Kenai River sockeye salmon have been integrated with long term research efforts by the Alaska Department of Fish and Game on these stocks. In addition, studies by the Limnology Laboratory and the fisheries development staff on Kodiak Island on these systems are included in data analysis. Study design and methodology builds off of earlier efforts. Planning and permitting of research activities and future rehabilitation efforts are coordinated through the USFWS Refuge staff in Soldotna.

## **PROPOSED PRINCIPAL INVESTIGATOR**

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## PERSONNEL

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Alaska Department of Fish and Game  
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## EMPLOYMENT:

October, 1991 to Present. Limnologist III, Principal Limnologist, FRED Division, Alaska Department of Fish and Game, Soldotna, AK. Responsibilities include establishing research objectives for the Statewide limnological investigations of the Commercial Fisheries Management and Development Division. This section provides direction for other components of the Division for determination of stocking rates for sockeye salmon in lakes and in the application of fertilization. This section also provides input to the commercial fisheries division for determination of the escapement goals for sockeye salmon. Supervise the limnology laboratory which completes water quality and plankton analysis for water samples taken from several hundred lakes statewide.

April, 1985 to October, 1991: Fishery Biologist IV, Regional Research Biologist, Westward Region, Alaska Department of Fish and Game. Responsible for establishing research objectives and priorities for the Westward Region Commercial Fisheries Division. This Division has management authority over extensive salmon and herring stocks on the Alaska Peninsula and Kodiak Island, in addition to management of the major shellfish stocks in the Gulf of Alaska and the Bering Sea. Annual ex-vessel value of these fisheries is several hundred million dollars. Research highlights included studies of crab larvae settling rates in the Gulf of Alaska and investigations on the effects of oil spill overescapement on the sockeye salmon production of major lakes on Kodiak Island.

May, 1982 to September, 1985 Acting F. B. IV, Susitna River Aquatic Studies Coordinator, Alaska Department of Fish and Game. The entire program under supervision included approximately 25 permanent and 50 seasonal employees. During this interim period, responsible for reorganizing the studies into a more efficient structure to meet the long term monitoring needs for determination of the effects of the Susitna project on the aquatic resources of the Susitna River. Supervised development of operational plans for 18 technical study programs on the Susitna River, assignment of priorities of tasks, and review of the technical merit of the programs proposed. Prior to January 1985. F. B. III, Resident and Juvenile Anadromous Project Leader, Su-Hydro Aquatic Studies Program, Alaska Department of Fish and Game. Supervised research programs on resident and juvenile anadromous fish in the Susitna River that may be impacted by development of the Su-Hydro Project. Technical studies included

development of models of sport fishery exploitation on arctic grayling populations, modeling instream flow responses of juvenile salmon habitat, development of baseline population parameters of resident fish and juvenile salmon and development of projections of supersaturated gas dissipation below the proposed dam sites.

January, 1981 to May, 1982: Fishery Biologist, Terrestrial Environmental Services, Anchorage, Alaska. Responsible for field and office review of the aquatic studies programs of the Alaska Power Authority for the Susitna Hydro-Electric Program. This responsibility included assisting the Alaska Department of Fish and Game in study plan development, providing preliminary assessment of impacts of the project on aquatic resources and presenting to the public progress of the aquatic studies programs.

May, 1980 to October 1980: Fishery Biologist, U.S. Fish and Wildlife Service, Soldotna, Alaska. Assisted on a radio-telemetry project and juvenile salmon habitat survey on the Kenai River, Six-mile Creek, and the Deshka River in the Cook Inlet area. Activities included tagging and radio tagging chinook and coho salmon, collection of juvenile salmon and measurements of associated habitat, and assisting in the analysis of scale patterns from Kenai River chinook salmon. Other activities included statistical analysis of data, report review, and preparation of a publication on the Kenai River chinook for Alaska magazine.

#### EDUCATION:

Ph.D. in Fisheries 1973

Major Field - Fisheries- Minor Field Pharmacology,  
Oregon State University, Corvallis, Oregon

M. S. in Biology, 1970 Major Field - Aquatic Biology Minor Field - Sanitary Biology,  
University of Utah, Salt Lake City, Utah

B. S. in Wildlife Biology, 1968, University of Montana, Missoula, Montana

Bruce P. Finney

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#### RELEVANT EXPERIENCE:

Stable isotopic studies of aquatic organic matter and carbonates.

Chemical and mineralogical analysis of geological samples.

Diatom analysis. Uranium-series isotope analysis and Quaternary

dating techniques. Analysis of sedimentary organic carbon, calcium carbonate and biogenic silica. Field experience in box, gravity and piston coring, hydrocasts and sediment trap deployment and recovery. Description and curation of geological samples. Computer-based statistical applications to geological data sets. Partitioning models of chemical composition using linear programming, factor analysis and chemical leaching studies. Time-series analysis and statistical testing.

EDUCATION:           University of Minnesota, 1979, B. S. Geology (with honors)  
                          Oregon State University, 1986, Ph.D. Geological Oceanography

Selected Publications:

Finney, B. P. and Johnson, T. C. (1991). Sedimentation in Lake Malawi (East Africa) during the past 10,000 years: a continuous paleoclimatic record from the southern tropics. *Palaeogeography, Palaeoclimatology, Palaeoecology*, 85: 351-366.

Finney, B. and Huh, C.A. (1989). History of metal pollution in the Southern California Bight: An update. *Environ. Sci. Technol.* 23: 294-303.

Finney, B., Lyle, M. and Heath, G.R. (1988). Sedimentation at MANOP Site H (Eastern equatorial Pacific) over the past 400,000 years: Climatically induced redox variations and their effects on transition metal cycling. *Paleoceanography* 3: 169-189.

Halfman, J. D., Johnson, T. C. and Finney, B. P. (1994). New AMS dates, stratigraphic correlations and decadal climatic cycles for the past 4 ka at Lake Turkana, Kenya. *Palaeogeography, Palaeoclimatology, Palaeoecology*, 111: 83-98.

Finney, B. P., Gardner, D. G. and Edwards, M. E. (1993). Late Quaternary climate change in Interior Alaska: Clues from reconstructing lake-level changes. *EOS*, 74: 332-333.

Finney, B. P. and McNeil, S. (1996). Sedimentary stable nitrogen isotopes in salmon systems: A tool for reconstructing long-term records of salmon abundance. Submitted to *Canadian Journal of Fisheries and Aquatic Sciences*.

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Prepared 4/8/97

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## EMPLOYMENT:

January 1993 - present: Biometrician for the Alaska Department of Fish and Game, Limnology Section, Commercial Fisheries Management and Develop Division, Soldotna, Alaska. Supervised by Dr. Dana Schmidt. Conduct statistical data analyses to evaluate factors that affect dynamics of the biota in lake ecosystems. Design limnological experiments and determine methods to estimate zooplankton and salmon abundance. Develop and approve methods to estimate hatchery contributions to the fishery. Develop, review, and conduct statistical analyses for projects related to the impact of oil on commercial fishery species. Provide biometrical consulting to area and regional biologists and statewide limnologists.

November 1991 - January 1993: Mathematical Statistician for the National Marine Fisheries Service, Auke Bay Laboratory, Juneau, Alaska. Supervised by Mr. Steven Ignell. Conduct statistical studies on community attributes of pelagic fauna in the north Pacific Ocean. Provide biometrical consulting, technical editing, and collaborative input on projects such as salmon bycatch and climate change studies.

January 1989 - May 1991: Statistics Teacher, Experimental Statistics Department, New Mexico State University, Las Cruces. Supervised by Dr. Michael Ames. Instruct laboratory courses in statistics for undergraduate science majors.

May - August 1990: Research Specialist (statistician), Department of Entomology, Plant Pathology, and Weed Science, New Mexico State University. Dr. Ellis Huddleston, Supervisor. Provide statistical modeling, analysis, and design of experiments related to agricultural field studies and pest management programs.

May - December 1988: Field Biologist, Biology Department, New Mexico State University. Supervised by Mr. Roger Skaggs. Conduct field population surveys and habitat analyses of night birds in Lincoln National Forest, New Mexico. Collect field data, supervise field personnel, and maintain data records. Develop operational strategies and conduct follow-up statistical estimation procedures.

August 1985 - June 1988: Graduate Assistant, Biology Department, New Mexico State University. Supervised by Dr. Ralph Raitt and Dr. Walt Whitford. Teach undergraduate biology and zoology laboratory courses. Collect data and maintain field ecology experiments for ecological research programs. Develop and conduct original field research on desert insect ecology.

June 1983 - May 1985: Research Specialist, Gordon Environmental Studies Laboratory, University of Montana, Missoula. Supervised by Dr. Philip Tourangeau. Manage data, conduct quality assurance/control procedures, and perform statistical analyses for

environmental science projects. Aid in the design and implementation of field research, primarily in the area of pollution biomonitoring.

#### EDUCATION:

- 1991 Master of Science, Experimental Statistics, New Mexico State University.
- 1988 Master of Science, Biology (ecology), New Mexico State University.
- 1983 Bachelor of Arts, Environmental Biology, University of Montana

Gary Kyle

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#### Experience:

April 1977 - April 1988: Project Biologist and later Area Biologist for the Division of Fisheries Rehabilitation, Enhancement, and Development of the ADF&G in Soldotna Alaska. Conducted and evaluated various fisheries enhancement and evaluation projects in the Cook Inlet watershed including limnological investigations of sockeye salmon producing lakes, and evaluation of hatchery stocking programs. Also, during the period I served as a project limnologist for the Limnology Section which involved the collection, analysis, and interpretation of limnological data from sockeye nursery lakes for assessment of rearing capacity and for modeling purposes.

April 1988 - present: Regional Limnologist for the Limnology Section for ADF&G in Soldotna, Alaska. Supervised by Dr. Dana Schmidt. As the Regional Limnologist for the Southcentral Region comprising of the Interior, PWS, Cook Inlet, and Alaska Peninsula; the primary purpose of this position is the supervision of staff in the coordination, assignment, prioritization, analysis, and review of subordinates work and interagency contract work related to lake fertilization and stocking projects, water quality monitoring projects, and fisheries and limnological research. In addition, the position is responsible for training subordinates, reporting and review of project results for publications and meetings, and administering state and non-state (contract) budgets.

#### Education:

1975 Bachelor of Science, Life Science/Natural Resources, University of Wisconsin.

#### Publications:

A total of 41 technical reports, 10 journal manuscripts, 26 formal presentations, and 6 magazine articles dealing with adult sockeye production, lake fertilization, lake stocking, and in-lake assessments of juvenile sockeye production.

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Alaska Department of Fish and Game  
Division of Sport Fisheries  
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## EXPERIENCE

### ALASKA DEPARTMENT OF FISH AND GAME, SOLDOTNA, ALASKA

Fishery Biologist III, October 1996- Present. Research Project Leader in Soldotna. This office is responsible for salmonid research projects on the Kenai Peninsula, including chinook salmon juvenile coded wire tag application and recovery projects, and resident salmonid (dolly Varden) distribution and life history studies.

Fishery Biologist II, June 1980- September 1996. Primarily responsible for Commercial Fisheries (CF) Division in-river adult sockeye salmon enumeration projects in Upper Cook Inlet (UCI). Also participated in a variety of habitat alteration evaluations including the Susitna Hydroelectric Project, the Chakachamna Hydroelectric and Diamond Chuitna Coal projects baseline environmental studies, state (DNR) land disposals and the proposed non-competitive coal leasing plans (DNR), the Mat-Su Borough land planning process (Susitna Area Plan), the Cook Inlet offshore mining lease program, the Matanuska Coal Field Competitive Lease and revisions to the Statewide Forestry Practices Act and the Mat/Su Valley Timber Harvest Economics Report.

Participated in projects to estimate the numbers of juvenile sockeye salmon rearing in Kenai and Skilak Lakes and the numbers of smolt migrating out of the drainage. The focus of the studies was expanded to provide information regarding the potential effects of high escapements (due to the Glacier Bay and Exxon Valdez oil spills) on system productivity. Developed and implemented a sockeye salmon productivity study of the Russian River and nine lakes in the Susitna River drainage.

### ENVIRONMENTAL RESOURCES AND TECHNOLOGY, INC., FORT COLLINS, COLORADO

Senior Entomologist, June 1973- January 1977. Responsible for terrestrial invertebrate research conducted on a variety of proposed energy development projects including Federal Oil Shale Tract leases in Northwest Colorado, an EPA study on the effects of sulfur dioxide and other coal combustion emissions on insects and insect-vegetation relationships in Colstrip, Montana, Newton, Illinois (coal-fired generator), Campbell County, Wyoming (coal gasification facility), Inola, Oklahoma (nuclear power facility), Iatan, Kansas (coal-fired generator) and Henderson, Minnesota (nuclear power facility).

## EDUCATION

BS in Biological Science, Colorado State University, Fort Collins, Colorado, 1973.

MS in Entomology, Colorado State University, Fort Collins, Colorado, 1974-1976,  
(thesis not completed)

## PUBLICATIONS

A total of 43 technical reports dealing with invertebrate and salmonid population studies.

## LITERATURE CITED

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IN REPLY REFER TO:

## United States Department of the Interior

## FISH AND WILDLIFE SERVICE

Kenai Fishery Resource Office

P.O. Box 1670

Kenai, Alaska 99811

Dana Schmidt  
Alaska Dept. of Fish & Game  
Division of Commercial Fisheries  
Management & Development  
34828 Kalifornsky Beach Road, Suite B  
Soldotna, Alaska 99669

April 3, 1997

Dear Dana:

I have reviewed your project proposal No. 97239 "Salmon carcasses and juvenile chinook salmon production in the Kenai River Ecosystem" and believe that it has the potential to improve sockeye salmon escapement management in the Kenai River. Presently, escapement levels are based upon a desired production level of fry and excess escapement is not considered beneficial or desirable in terms of production. If successful, your project would provide a direct link between sockeye salmon carcass nutrients and juvenile chinook salmon production in the Kenai River system. This information would be very beneficial in determining sockeye salmon escapement levels which insure sustainability of both sockeye and chinook salmon populations in the Kenai River system. This multi-species approach is indeed an ecosystem approach to management of Kenai River salmon resources.

Sincerely,

Gary M. Sonnevil  
Project Leader

**1997 EXXON VALDEZ TRUSTE COUNCIL PROJECT BUDGET**

October 1, 1996 - September 30, 1997

<b>Budget Category:</b>	<b>Authorized FFY 1996</b>	<b>Proposed FFY 1997</b>						
Personnel		\$101.4						
Travel		\$2.2						
Contractual		\$39.2						
Commodities		\$5.8						
Equipment		\$0.0	<b>LONG RANGE FUNDING REQUIREMENTS</b>					
Subtotal	\$0.0	\$148.6	Estimated FFY 1999	Estimated FFY 2000	Estimated FFY 2001	Estimated FFY 2002	Estimated FFY 2003	
General Administration	\$0.0	\$18.0						
Project Total	\$0.0	\$166.6	\$100.0	\$0.0	\$0.0	\$0.0	\$0.0	
Full-time Equivalents (FTE)		1.8						
Dollar amounts are shown in thousands of dollars.								
Other Resources								
<p><b>Comments:</b></p> <p>This project description has been revised from the 1997 submittal to reflect reasoning for limiting the program to Chinook salmon. Expansion to other species may be warranted in following years, depending upon results of the proposed investigation. Based on technical discussions with Bilby et al. (1996), we have expanded the study to include a non-anadromous control stream and will use juvenile rainbow trout to examine baseline marine isotope values.</p> <p>This project will provide for an initial year of field data collection of the different trophic levels of the Kenai River mainstem ecosystem and selected tributaries. These data will be used to complement existing work on the lake ecosystems, to understand the role of carcass nutrients in maintaining current levels of productivity in the Kenai River.</p> <p>This FY98 budget submittal reflects a logical extension of the overescapement studies on the Kenai River. This program, however, is not dependent upon data or analysis collected in 97258, although the program was developed based on observations and analysis conducted under the overescapement program. The budget submitted reflects manpower contributions from two divisions of ADF&amp;G, who will work cooperatively in completing this project. The stable isotope analysis and other technical assistance will be provided through Dr. Bruce Finney of the University of Alaska in Fairbanks through a non-competitive RSA. The project is planned for two years, with the first year reflecting detailed study design development and data collection. FFY99 will entail sample processing, data analysis and report writing.</p>								

**FY98**

Project Number: 98239  
 Project Title: Kenai Salmon Carcass Nutrient Investigation  
 Agency: ADF&G

**FORM 3A  
 TRUSTEE  
 AGENCY  
 SUMMARY**

Prepared: April 15, 1997

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<p><b>FY98</b></p>	<p>Project Number: 98239          Project Title: Kenai Salmon Carcass Nutrient Investigation          Agency: ADF&amp;G</p>	<p>FORM 3B          Personnel          &amp; Travel          DETAIL</p>
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Project Number: 98239  
Project Title: Kenai Salmon Carcass Nutrient Investigation  
Agency: ADF&G

FORM 3B  
Personnel  
& Travel  
DETAIL

2 of 4

'9/97

**1997 EXXON VALDEZ TRUSTEE NCIL PROJECT BUDGET**

October 1, 1996 - September 30, 1997

<b>Contractual Costs:</b>		Proposed
Description		FFY 1997
Rept	Long distance telephone and toll costs 10 months @ \$ .1, copying and binding- 50 copies @ \$.01)	\$1.5
Rept	Computer equipment repair (1 hard drives @ \$.5, one motherboard @ \$1.0)	\$1.5
Rept	Postage (10 months @ \$.05), photo processing (5 rolls @ \$.02), messenger service (\$.1)	\$0.7
Field	Film processing and purchasing	\$0.1
	Vehicle repair	\$0.2
Field	Auto equip & parts	\$0.2
RSA	Contract for stable isotope analysis- University of Alaska IMS \$0.1/sample x 350 samples	\$35.0
When a non-trustee organization is used, the form 4A is required.		
<b>Contractual Total</b>		<b>\$39.2</b>
<b>Commodities Costs:</b>		Proposed
Description		FFY 1997
Rept	Office supplies-Paper (\$.25), Xerox supplies and computer printer supplies (\$.25)	\$0.0
Rept	Laboratory glassware (\$.5), chemical reagents (\$1.0)	\$1.5
	Field sampling gear, invertebrate nets (\$.25) backpack shocker repair, parts (\$.5)	\$0.8
Rept	Photographic supplies - camera parts and film	\$0.1
x	Stationery (\$.2) and duplicating supplies - Xerox toner-supplies (\$.4)	\$0.0
Field	Raingear, hip boots and gloves for 4 people @ \$.25	\$1.0
Field	Food (60 marl days @ \$.015/day)	\$0.9
Field	Flotation coats for 2 people @ \$.25	\$0.5
Field	Fuel costs- all vehicles and vessels	\$1.0
<b>Commodities Total</b>		<b>\$5.8</b>

**FY98**

Project Number: 98239  
 Project Title: Kenai Salmon Carcass Nutrient Investigation  
 Agency: ADF&G

**FORM 3B**  
 Contractual &  
 Commodities  
 DETAIL

Prepared: April 15, 1997

October 1, 1996 - September 30, 1997

**FY98**

FORM 3B  
Equipment  
DETAIL

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4/9/97