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Investigations of disease factors affecting declines of Pacific herring populations ir Prince William Sound, AK

Project number:	9616: 9616:	2 (was 953205)	
Restoration Category:	Research and Monitoring		
Lead Agency:	Alaska Department of Fish and Game		
Proposer:	Univ. of Washington. U.C. Davis, National Biological Service &		
	Simon Fraser Univ.		
Cooperating Agencies:	National Biological Service (NBS), Seattle, WA		
Duration:	3 years (FY-96 thru FY-98)		
Cost of project:	FY 96 G FY 97 2 FY 98 2	635.0 510.6 461.7	
Total	1150		
Geographic area: Injured resource:	Prince William Sound Herring	d, Sitka Sound, AK	

ABSTRACT

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Field and laboratory studies will focus on Viral Hemorrhagic Septicemia Virus (VHSV) and *lchthyophonus hoferi*, a pathogenic fungus, to determine their role in the disease(s) and mortality observed in Prince William Sound herring since 1993. Herring in PWS will be monitored three times per year for signs of disease and immune status. Specific Pathogen-Free herring will be used to determine the degree of mortality, blood chemical changes and pathogenicity produced by these organisms alone and in combination with exposure to stressors such as petroleum hydrocarbons, temperature and crowding.

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INTRODUCTION

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In 1993, over half of the 135,000 tons of spawning Pacific herring expected to return to PWS failed to appear. Among those that that did return, 15-42% behaved abnormally and had hemorrhages beneath the skin. Pathologists from ADF&G isolated VHSV from these herring and from skin lesions of a Pacific cod caught nearby. At the same time, herring with similar skin lesions were found near Kodiak Island, although the fishery there met predicted expectations. In 1994 only 20,000 tons of herring returned to PWS and little or no spawning occurred. In 1994 20% of spawning fish had moderate or severe external lesions. VHSV was isolated from 11/233 (5.7%), and 62/212 (29%) had *lchthyophonus*. Samples are currently being taken in PWS as well as Sitka Sound to determine the role of VHSV in the etiology of the 1993 - 94 epizootics. By comparison, prevalence of *lchthyophonus* in PWS herring from 1989 through 1992 was never more than 15%; hence it was considered a possible significant cause of morbidity in 1994, but the initiating cause of the population declines before 1993 spawning remains unknown.

This project consists of three components: 1) Field monitoring, 2) Laboratory disease and stressor evaluation and 3) Biochemical and physiological changes. The study is designed to determine whether VHSV or I. hoferi are responsible for the herring mortality and lesions observed in Prince William Sound since 1993, and to monitor their recovery and identify biomarkers which would indicate the presence of disease organisms. It will also examine the possibility that exposure of herring to crude oil could reduce their resistance to infection by pathogenic organisms. The project began in 1995 (95320S) with on-site monitoring and the production of specific pathogen-free (SPF) herring for disease-stressor interaction studies. Monitoring is continuing in PWS on pre and post-spawning herring as well as late summer adults. Embryos from Prince William Sound herring are also being incubated in filtered and u.v. sterilized seawater in order to produce SPF larvae. As these eggs hatch and the larvae grow to appropriate size and age, they will be exposed to both VHS virus and *I. hoferi*, alone and in conjunction with exposure to petroleum hydrocarbons. Following these exposures the herring will be examined for survival, gross and microscopic lesions (disease), behavioral changes and ultimately reproductive success. In addition to exposure to pathogens and chemical stressors, herring will also be subjected to crowding conditions and temperature extremes to determine if physical stresses could be partially responsible for the observed disease and mortality. Blood chemical measurements will be done on wild and laboratory reared herring to determine whether exposure to the various pathogens alters normal physiologic functions and whether biomarkers could be identified which would aid in future identification of similar disease problems.

NEED FOR THE PROJECT

A. Statement of Problem

Pacific herring (*Clupea pallasi*) are an injured biological resource in Prince William Sound (PWS) classified as "not recovering". Because of the population declines in 1993 and 1994, commercial herring fishing was closed in both seasons, resulting in economic losses and lost services. The fishery is expected to be closed again in 1995. Following the population declines in herring, there have also been significant declines in marine birds and mammals which depend on herring as a forage food. Thus, the reduction in herring numbers in PWS has the potential for significant impacts throughout the ecosystem. Pacific herring are also a major subsistence and economic resource in Prince William Sound. 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 Chenega and Tatitlek. There are also five commercial herring fisheries in PWS. The ex-vessel value of the herring fisheries in 1992 was \$12.0 M and the average annual value for the previous 10 years was \$8.3 M. In 1993, the ex-vessel value dropped to \$2.0 M due to low abundance and the prevalence of small fish with low market value. As of the spawning season of 1995, there was no indication of recovery of this species.

B. Rationale

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Following the *Exxon Valdez* oil spill (EVOS) in 1989 the Alaska Department of Fish and Game (ADF&G) conduced studies on Pacific herring in Prince William Sound. Following these studies the the Herring Research Synthesis group reported to the Trustee Council that Prudhoe Bay crude oil did cause damage to herring at the whole animal, genetic and biochemical level. The group also predicted that the most severely impacted age groups would be the 1988 and 1989 year classes which would enter the spawning population in 1992 and 1993. Based on its findings, and the available scientific literature, the group predicted that damage to the herring's immune system could result in severe disease outbreaks and possible neoplasia in subsequent years. By 1993 herring were considered "recovered" and no herring studies were conducted in Prince William Sound. However, a massive die-off occurred, as predicted, accompanied by the presence of viral hemorrhagic septicemia virus (VHSV) and ultimately over 75% of the spawning population was lost.

Identification of the organisms responsible for herring mortality, and the conditions associated with the observed diseases is critical to successful management. Constant seasonal monitoring of the disease status of herring will determine whether the disease(s) is abating, or if losses continue. Studies on the specific causes of the observed lesions will identify the organisms and conditions causing the loss of herring. If herring were damaged as the result of exposure to crude oil or its components, it is important to determine if the damage is short term or permanent. Short term damage could have produced the high level of mortality observed since 1992, but recovery would be relatively rapid once unaffected fish (eg. post-spill year classes) begin to dominate the spawning biomass. If however, the damage is more permanent (eg. heritable), it could take much longer for the pathogen(s) and hosts to develop a benign relationship compatible with long term co-existence without high mortality rates. Biochemical and physiologic studies will identify biomarkers indicative of the presence of disease(s) and enable managers to recognize potential problems before they occur.

During the recovery period management practices can be used to protect severely depleted spawning stocks during recovery. Selective harvesting of specific year classes might also be used to speed recovery. It is also important to avoid crowding herring into confined areas where transmission would be increased, thus producing a pool of infected individuals which could transmit the pathogens to uninfected individuals. It is also important to devise management practices which prevent inadvertent transport of potentially virulent strains of the pathogen(s) to other herring populations. Sanitizing vessels and equipment between fishing sites would prevent the spread of disease from one population to another.

Considerable research is needed to determine the role of VHSV, *I. hoferi* and possibly other organisms in the precipitous decline of the herring stock in Prince William Sound. The role of chemicals (PAH, alkanes, etc) and environmental factors on disease resistance should also be examined. This will require field surveys of the distribution of pathogens as well as experimental infections to fulfil Koch's postulates. and controlled exposures to chemical

stressors to determine the role of petroleum hydrocarbons on the disease resistance of herring.

C. Summary of Major Hypotheses and Objectives (FY 96 - 98)

<u>Hypotheses</u>:

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- a) VHS virus is the cause of lesions and/or mortality observed in Pacific herring in PWS.
- b) Exposure to stressors can decrease the immune resistance of herring to VHS virus.
- c) Ichthyophonus hoferi is pathogenic (eg. cause disease) in Pacific herring.
- d) Exposure of herring to stressors can decrease their resistance to infection by *l. hoferi*.
- e) The combination of infection by VHSV or *I. hoferi* and stressors can cause morbidity and mortality in Pacific herring in excess of what would occur if they were exposed singly.
- Herring populations will begin to recover when the frequency of infection is reduced to pre-1993 levels.

Objectives:

<u>FY 96</u>

- 1. Investigate the impact of disease on herring population size and age structure.
- 2. Determine the relationship between organisms and lesions, plasma chemistry and immune status.
- 3. Determine the role of reproductive stage on herring health.
- 4. Establish SPF herring in the laboratory for use in definitive disease studies on VHSV and Ichthyophonus hoferi
- 5. Fulfil Koch's Postulates for VHSV and I. hoferi in SPF Pacific herring.
- 6. Establish an SPF model system for studying VHSV and *I. hoferi* under controlled conditions.

D. Completion date

September 30. 1998

COMMUNITY INVOLVEMENT

An annual progress report will be presented at a Restoration Science Workshop to be held in Anchorage each January. Principal investigators will be available to speak with the media and public while actively working in PWS and by phone during the remainder of the year. Fishermen interested in learning more about disease identification and sanitizing vessels and equipment can contact the principle investigators.

FY 96 BUDGET

Personnel	-203.8	34,2
Travel	-21.8	8,0
Contractual	-162.0	549.2
Commodities	~20.5	15.0
Equipment	-4.1	0.0
Operating fees	-6.8	
Subtotal	419.0	606.4
Indirect costs	<u>91.8</u>	28.6
Total	510.8	635.0

SECTION I

Field Component

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PROJECT DESIGN, Field Component (University of California, Davis)

A. **Objectives**

The field component of this proposal has three objectives to help determine why herring populations are not recovering in PWS:

- 1. Determine which pathogens cause lesions and determine the relation among Viral Hemorrhagic Septicemia virus (VHSV), *Ichthyophonus*, macroscopic and microscopic lesions, plasma chemistries, and immune status.
- 2. Determine the role of reproductive stage on the general health of herring. Are lesions, *Ichthyophonus*, and VHSV more severe during a given reproductive stage?
- 3. Investigate the impact of disease on herring population size and structure of herring. Are fish of a particular year class more likely to be diseased than fish of other year classes? Does a history of previous oil exposure correlate with prevalence and severity of disease?

B. Methods

* N.J There is yet no indication that the importance of disease in the decline of PWS herring has diminished. Therefore, field sampling to continue to document the dynamics of this epizootic is a high priority of the project. The most important pathogen contributing to morbidity of Pacific herring in 1993 was thought to be VHSV, whereas *Ichthyophonus* was thought to be most important in 1994. Both diseases involve multiple organs, and interaction with other parasites and lesions must be explored. Further, a new disease may emerge as most important in 1995 and beyond. Parasites are a normal component of wild fish populations, but under conditions of stress, parasite pathogenicity can increase. Ancillary studies such and immune function as plasma chemistries are needed to determine the effect of parasites on fish health.

In order to test the hypothesis that a given disease is significant, complete histopathology is required. Our basic assumption is that VHSV and *Ichthyophonus* are important pathogens, but that the full cause of population decline is unknown and may involve other pathogens through direct or synergistic effects. To determine the role of disease, we propose intensive examination of relatively few fish as opposed to cursory examination of many fish. For example, in 1994 we learned that moderate to severe external lesions were fairly good indicators of VHSV infection but were relatively poor indicators of *Ichthyophonus* status. External examination takes about 20 seconds per fish (i.e., examination of many fish would be inexpensive), but limiting examination to external lesions in 1994 would have failed to identify about one half of the sick fish in the population.

To test the hypothesis that reproductive stage affects pathogenesis, sampling is needed during prespawning, spawning, and postspawning (Fall), and during the period of gonadal development and peak condition (Fall). To test the hypothesis that fish that were yearlings at the time of the spill (i.e., 1988 year class) are more susceptible to disease than are other year classes, a minimum sample size of 300 would be ideal (Fritz Funk, ADFG, personal communication). In 1994, 233 fish were sampled and the age structure was similar to more extensive age-weight-length measurements taken by ADFG

on 450 fish. Therefore, we propose sampling 240 fish in the Spring for age analysis; smaller year classes will be combined to increase statistical power of age-specific analyses, if needed. Data from the 80 fish sampled in the Fall will be used to compare population disease prevalence between and within sites. A sample size of 80 is sufficient to have 95% confidence that disease with a prevalence \geq 4% will be detected in at least one fish sampled (Becker and Grieb 1987). Power analysis cannot be done for determining sample size for comparing Sitka Sound and PWS because no baseline data are available from Sitka Sound. After results from the first year of comparative study are available, sample size for the second year of comparative study might be changed to increase power of statistical tests.

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To best characterize the condition of herring in Prince William Sound and Sitka Sound, herring will be subjected to complete necropsy using the following sampling schedule (as field conditions allow) over the course of three years of study:

Dates	Location	Reproductive Stage	Number of Fish
FY96:			
early Oct., 1995 (4 days)	Sitka Sound	peak condition/ gonadal development	80
mid-Oct., 1995 (4 days)	Prince William Sound	peak condition/ gonadal development	80
mid-late March, 1996 (3 days)	Sitka Sound	prespawning	80
late March/early April, 1996 (3 days)	Sitka Sound	spawning/post-spawning	160
early-mid April, 1996 (3 days)	Prince William Sound	prespawning	80
mid-late April, 1996 (3 days)	Prince William Sound	spawning/post-spawning	180
		Total Fish, FY96:	660
FY97:			
early Oct., 1996 (4 days)	Sitka Sound	peak condition/ gonadal development	80
mid-Oct., 1996 (4 days)	Prince William Sound	peak condition/ gonadal development	80
early-mid April, 1997 (3 days)	Prince William Sound	prespawning	80
mid-late April, 1997 (3 days)	Prince William Sound	spawning/post-spawning	180

_	Dates	Location	Reproductive Stage	Number of Fish
			Total Fish, FY97:	420
-	FY98:			
	mid-Oct., 1997 (4 days)	Prince William Sound	peak condition/ gonadal development	80
	early-mid April, 1998 (3 days)	Prince William Sound	prespawning	80
	mid-late April, 1998 (3 days)	Prince William Sound	spawning/post-spawning	180
			Total Fish, FY98:	340
•			Total Fish, 3-year study:	1420

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Herring will be sampled by gill net, purse seine, or cast net. To minimize effects of capture and holding, fish will be held no longer than four hours before necropsy during spring sampling. For Fall samples, herring will be captured by purse seine daily and held in large containers (10 L of water per fish). If fish cannot be captured alive in the Fall, then plasma chemistries will be eliminated from the analysis at a savings to the project. In PWS, necropsies will be done on anesthetized fish on a chartered vessel. The vessel will accompany the R/V *Montague* and chartered catch-vessel during Fall hydroacoustic surveys that are proposed as part of project 96320-N (hydroacoustics) and 96166 (natal habitats). In Sitka, Spring samples will be necropsied in an AK Dept. of Fish and Game garage in Sitka. Sitka does not normally have a Fall hydroacoustic survey. Therefore, a vessel equipped with hydroacoustic gear will be chartered to find fish; after fish are found, the R/V *Medea*, a large research vessel, will be used to catch the fish and for on-vessel necropsy.

During necropsy, each fish will be anesthetized in tricaine methane sulfonate (Finquel®) and visually screened for external lesions, which are ranked as none (0), mild (1), moderate (2), or severe (3). Measurements on each fish include body weight, standard length, age (from scales), liver weight, and gonad weight. Otoliths are archived for later use if information on annual growth rates is desired. Samples will be taken for several types of analysis (listed in order of priority):

- a. Histopathology (fix in 10% neutral buffered formalin) gill, spleen, liver, gonad, heart, stomach, intestinal tract, exocrine pancreas, kidney, skeletal muscle, skin, brain, and other gross lesions. All tissues will be examined for lesions, which are scored as described for gross lesions and using the type specimens developed in 1994. Oocyte stages will be quantified by counting a representative sample on the slide prepared on histopathology. Also, a touch prep of anterior kidney from each fish is made on a glass slide, stained, and examined for the myxosporean *Ortholinea orientalis*. Histopathology will be done under the direction of Dr. Gary Marty at the University of California, Davis.
- b. Virus isolation (put in plastic bags, on ice) anterior kidney, spleen, and any severe skin lesions. Although VHSV grows well on non-herring cell lines, other viruses might not. A cell

line derived from Pacific herring will be used to attempt isolation of other, yet unknown viruses. Virus isolation will be done under the direction of Dr. Ted Meyers at the ADFG Fish Pathology in Juneau (Meyers et al. 1994).

- c. Hematology blood will be drawn from the caudal vein into a Lithium-heparinized syringe. Packed cell volume (PCV) is determined on site. A smear is made for analysis of erythrocyte morphology (for diagnosis of Viral Erythrocytic Necrosis) and for white blood cell differential counts. Plasma is refrigerated for no more than 72 h, or frozen, for analysis of osmolality, total protein, albumin, cholesterol, glucose, total bilirubin, ALP, ALT, AST, CPK, GGT, sodium, potassium, chloride, phosphate, bicarbonate, lactate, and calcium. Determination of osmolality requires 50 µL of sample, to be analyzed on a Micro Osmometer Model 3MO-plus from Advanced Instruments (Norwood, MA). All other analytes can be done with 200 µL of sample using a Monarch-plus analyzer from Instrumentation Laboratories. To minimize protein denaturation, all enzyme levels are determined at 25° C. Dr. Chris Kennedy at Simon Fraser University will oversee screening of blood smears for Viral Erythrocytic Necrosis and will perform white blood cell differential counts. Other samples will be archived for later analysis, if warranted.
- d. Bacteriology for each fish with severe gross lesions, a sterile loop is stabbed into the anterior kidney and then streaked on Trypticase Soy Agar (TSA) and Marine agar for bacterial isolation. Ulcers will be preserved for histopathology and virology, but they will not be cultured (superficial bacteria can be diagnosed on histopathology).
- e. Immunology As a basic measure of immune status, differential leukocyte counts will be done on blood smears (under the direction of Dr. Christopher Kennedy, Simon Fraser University). Absolute leukocyte numbers will be estimated from the smear. Other immune function tests have not previously been developed for Pacific herring, but an ELISA assay specific for herring IgM is being developed in FY95 (95320S), with analysis to begin in FY96. A 100-µL sample of plasma from each fish will be frozen and later analyzed for immunoglobulins. Lymphocyte mitogen stimulation assays were considered, but special needs of the assay (e.g., sterile collection of cell suspensions) were determined to be too great for conditions on vessels available for this project. Plasma cortisol values have been shown to rise in other species within minutes of capture (capture stress); because herring will be held up to 4 h before necropsy, and cortisol determination is not readily automated, cortisol determinations will not be done on field-caught samples.
- f. Body condition A wedge of dorsal body musculature is removed from just caudal to the operculum of each fish and frozen in a 1.5-mL Eppendorf tube. Stable isotope analysis will be done only if indicated by other results.
- g. Cytochrome P450 induction Liver (0.1-0.2 g) is frozen and archived in 1.5-mL Eppendorf tubes. Analysis will be done only if indicated by results from virus isolation, histopathology, and hematology. Liver will not be archived if total liver weight is less than 0.4 g (e.g., from small fish).

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h. Age, weight, and length (AWL) measurements - Additional herring (to total 450 per sample period at each site) will be sampled for age, body weight, standard length, and gonad weight. These additional fish will not be subjected to complete necropsy or be examined by the pathologists.

The ADFG fisheries laboratory in Cordova, Alaska, will catch fish for necropsy, collect age and length data, prepare formalin and containers for tissue fixation, provide data recorders for each pathologist on site, and ship all samples.

Results from virus isolation will be reported as a VHSV titer. Results from analysis for Viral Erythrocytic Necrosis (blood smear) and histopathologic analysis will be reported for each lesion, and semiquantitatively ranked on a four-point scale (0,1,2, or 3) as described for gross lesions. Results from immunoglobulin ELISA assays will be reported as percent absorbance.

This study is designed to diagnose any type of disease that is causing morbidity in herring. Results will be compared with previous years of study. The following table lists Prevalence (%) of parasites and virus in adult Pacific herring in Prince William Sound, Alaska, 1989-1994:

Sample Date	Goussia clupearum	Ichthyophonus hoferi	Ortholinea orientalis	Viral Hemorrhagic Septicemia virus
1989 April (n = 40)	63	13	TNE ^a	TNE
1990 October (n = 99)	60	15	6.1	TNE
1991 April (n = 59)	54	5.1	1 7	TNE
1991 October (n = 48)	54	2.1	15	TNE
1992 April (n = 105)	53	5.7	3.1	TNE
1993 April	NR⁵	NR	NR	2 of 3 5-fish pools
1994 April (n = 212)	61	29	5.7	4.7 (n = 233)

*TNE = Tissue not examined

 $^{b}NR = not reported$

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Samples from 1994 had several other parasites, but in previous years appropriate tissues for comparisons were not examined. In order of decreasing prevalence, other parasites in 1994 samples included: (1) Anisakidae in the peritoneal cavity, 100%; (2) intestinal coccidian (*Goussia* sp. ?); 91%;

(3) gall bladder myxosporean Ceratomyxa auerbachi, 19%; (4) branchial monogenetic trematodes Gyrodactylus spp, 13%; (5) branchial ciliated protozoans, mostly Trichodina spp., 12%; (6) renal intratubular ciliated protozoan, species unidentified, 11%; (7) branchial Epitheliocystis, 10%; (8) gastric intraluminal trematodes, e.g., Hemiuridae, 8.6%; and (9), intestinal trematodes, e.g., Lecithaster gibbosus, 5.7%. Proposed for 1996 through 1998, prevalence of these parasites will again be determined, and associated lesions and alteration in plasma chemistries will be described. Study in 1994 found little association between parasites and disease except for Ichthyophonus and VHSV.

Several lesions and other observations will be scored for each organ; listed below are those lesions and significant findings from 1994 samples:

- Brain: Ichthyophonus, meningeal eosinophilic granular leukocytes, and granulomatous meningitis; in 1994, prevalence of Ichthyophonus was 8.0%, brain Ichthyophonus was the best marker of increased plasma creatine phosphokinase (CPK), and granulomatous meningitis was nine times more likely in VHSV(+) fish than in VHSV(-) fish.
- Gall bladder: intraluminal myxosporean (*Ceratomyxa auerbachi*); in 1994, prevalence of *Ceratomyxa* auerbachi was 19%, but the parasite was not associated with alterations in plasma chemistries. Examination of the gall bladder is included with the liver (i.e., no extra expense for analysis).
- Gill (for purposes of this study, the gill is composed of arches, filaments, and lamellae): Ichthyophonus, gill arch inflammation and/or hematopoiesis, lamellar hyperplasia, monogenetic trematodes (e.g., Gyrodactylus spp.), foreign body granulomas, Epitheliocystis, and ciliated protozoans (e.g., Trichodina spp.); in 1994, prevalence of Ichthyophonus was 13%, branchial trematodes were increased in fish with moderate to severe external lesions, and gill arch inflammation was six times more likely in VHSV(+) fish than in VHSV(-) fish.
- Gonad: Ichthyophonus, eosinophilic granular leukocytes, focal granulomatous inflammation, pigmented macrophage aggregates, seminiferous tubule distension (male only), hyalinized vessel walls (female only); oocyte stage (yolked and non-yolked eggs); in 1994, prevalence of Ichthyophonus was only 1.4%, and other lesions were minimal. However, several plasma chemistries were significantly correlated with gonad weight.
- Gross Lesions: caudal fin fraying, caudal fin reddening, fin base reddening, focal skin reddening, diffuse skin reddening, iris reddening, number of peritoneal Anisakidae, and gonadal fullness.
- Heart: Ichthyophonus, atrial phagocyte hypertrophy, myocardial mineralization, thrombosis, epicarditis, and focal parenchymal leukocytes; in 1994, prevalence of Ichthyophonus was 18%, and myocardial mineralization was 27 times more likely in VHSV(+) fish than in VHSV(-) fish.
- Intestine: Ichthyophonus, arteriolar focal intimal hyperplasia, foreign body granuloma, submucosal eosinophilic granular leukocytes, Anisakidae, steatitis, intestinal coccidian (Goussia sp.?), and intraluminal trematode (e.g., Lecithaster gibbosus); in 1994, prevalence of Ichthyophonus was 8.5%, and focal arteriolar intimal hyperplasia was 5 times more likely in VHSV(+) fish than in VHSV(-) fish. Further, a new coccidian parasite (Goussia sp.?) was in 91% of intestinal

sections, but the parasite was not associated with significant alterations in plasma chemistries.

Kidney (trunk): pigmented macrophage aggregates, granulomatous inflammation, *Ichthyophonus*, hematopoietic cells (relative area/volume), congestion, intratubular mineral, tubular epithelial vacuolation, tubular dilation, intratubular protozoan (probably ciliates), interstitial cell necrosis, and intratubular myxosporean (*Ortholinea orientalis*); in 1994, prevalence of *Ichthyophonus* was 20%, kidney was the best marker of increased plasma aspartate aminotransferase (AST), and the myxosporean *Ortholinea orientalis* was associated with renal granulomatous inflammation and increased scores for renal macrophage aggregates.

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- Liver: hepatocellular glycogen depletion, pigmented macrophage aggregates, granulomatous inflammation, *Ichthyophonus*, eosinophilic granular leukocytes (in perivascular or pericholangial connective tissue), lipidosis, focal parenchymal leukocytes, *Goussia* [Eimeria] *clupearum*, focal necrosis, single cell necrosis, and cholangitis/biliary hyperplasia; in 1994, prevalence of *Ichthyophonus* was 16%, and focal hepatic necrosis was 11 times more likely in VHSV(+) fish than in VHSV(-) fish. Several lesions were associated with changes in plasma chemistries. Further, 13% of the fish had abundant sporulating oocysts of *Goussia clupearum*, but these parasites were not associated with increased plasma chemistries.
- Pancreas (exocrine): pigmented macrophage aggregates, and zymogen granule depletion; in 1994, all fish with severe external lesions had severe zymogen granule depletion,
- Skin and Skeletal muscle: Ichthyophonus, myodegeneration and necrosis, perivascular leukocytes, myositis, and arteriolar focal intimal hyperplasia.; in 1994, prevalence of Ichthyophonus was 18%, but most lesions were mild. Ulcers had filamentous bacteria: a possible third cause of mortality (after Ichthyophonus and VHSV).
- Spleen: congestion, pigmented macrophage aggregates, granulomatous inflammation, *Ichthyophonus*, ellipsoid hyperplasia/hypertrophy, and arteriolar focal intimal hyperplasia; in 1994, prevalence of *Ichthyophonus* was 18%, and several lesions were associated with alterations in plasma chemistries. Further, splenic congestion was a biomarker of poor egg viability in herring sampled from Rocky Bay in PWS in 1992 (Kocan et al. In review).
- Stomach: Ichthyophonus, foreign body granuloma, submucosal eosinophilic granular leukocytes, serositis, intraluminal trematodes (e.g., Hemiuridae), and focal parenchymal leukocytes; in 1994, prevalence of Ichthyophonus was 10%, and severe infiltrations of gastric submucosal eosinophilic granular leukocytes were nine times more likely in VHSV(+) fish than in VHSV(-) fish.

Quality control and quality assurance are part of all examinations. For necropsy examination, two pathologists are on site at all times; when questionable or difficult lesions are encountered, the second pathologist is consulted. For histopathology, sections are coded for blind study; all 500 Spring samples will be coded as one group, and the 160 Fall samples will be coded as a second group. Tissues from each fish are assigned a random number and tissues are examined in ascending numerical order. The first 15 specimens of each organ are examined by two pathologists and lesions are scored independently. The pathologists then compare scores and modify diagnoses as necessary to

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come to a consensus. One pathologist then scores all specimens of a given organ and assigns type specimens for later review. To maximize comparability of results to 1994 and 1995 results (94320S and 95320S), type specimens described for the 1994 data will provide the basis for diagnoses in 1996 and beyond. The University of California, Davis, has three pathologists available to read sections and a fourth pathologist available for review; in the event of personnel change, remaining pathologists will increase effort on this project.

Type specimens developed on samples in 1994 (94320S) will be followed whenever possible; examples from the liver follow:

- I. Atly = Autolysis (a check for adequacy of fixation). Changes in membrane integrity begin immediately after death.
 - A. score = 0; no membrane changes, erythrocytes stained intensely (type specimen = 94H74-1B).
 - B. score = 1; loss of membrane integrity; hepatocytes had fragmented nuclei and pale basophilic cytoplasm; changes were probably due to autodigestion from leakage of bile (type specimen = 94H74-73B).
 - C. score = 2; none were moderate.
 - D. score = 3; none were severe.

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- II. Art = Artifact. Tissue changes that were not inherent in the tissue sampled. Sources of artifact included handling at necropsy, processing, sectioning, and staining. Artifact is scored on the basis that it impedes interpretation of tissue morphology. Examples of artifact include splits, bubbles, or knife marks in tissues.
 - A. score = 0; sections had no tissue alterations that would impede analysis or photography of any part of the sections (type specimen = none).
 - B. score = 1; tissue alterations were present, but most areas could still be photographed without artifact, and analysis for lesions was unaffected (type specimen = 94H74-1B).
 - C. score = 2; tissue alteration prevented analysis for lesions in some areas and photography would be unacceptable anywhere (type specimen = 94H74-159B).
 - D. score = 3; tissue alterations were too extensive for histopathologic analysis (type specimen = none were severe).
- III. GD = glycogen depletion. A lesion in hepatocytes; hepatocytes normally have abundant cytoplasmic glycogen stores characterized by a large volume of clear, irregular, poorly demarcated vacuoles (= glycogen vacuoles).
 - A. score = 0; hepatocytes had abundant glycogen vacuoles (type specimen = none).
 - B. score = 1; glycogen vacuoles were smaller, but still larger than nuclei (type specimen = none).
 - C. score = 2; glycogen vacuoles were smaller than or about equal to nuclear diameter (type specimen = 94H74-163B).
 - D. score = 3; glycogen vacuoles were absent for most hepatocytes (type specimen = 94H74-62B).
- IV. LMA = liver macrophage aggregates. A lesion in the hepatic stroma or capsule. Macrophage aggregates were pigmented yellow-brown to green-brown, and occasionally contained

lymphocytes.

- A. score = 0; no macrophage aggregates (type specimen = none).
- B. score = 1; sections had <7 MAs greater than 60 μ m in diameter per 100X field (type specimen = 94H74-4B).
- C. score = 2; sections had ≥7 but <14 MAs greater than 60 µm in diameter per 100X field (type specimen = 94H74-1B).
- D. score = 3; sections had ≥14 MAs greater than 60 μm in diameter per 100X field (type specimen = 94H74-14B).
- V. LGR = liver/hepatic granulomas (or focal granulomatous inflammation). Focal hepatic granulomatous inflammation, composed of nonpigmented macrophages, was distributed throughout the parenchyma, commonly associated with portal tracts. Often, nonpigmented macrophages expanded pre-existing LMAs (e.g., 94H74-141B). As with LMAs, LGRs occasionally contained eosinophilic granular leukocytes (EGLs). Cytoplasmic staining in granulomas varied from mostly eosinophilic (94H74-127B) to mostly basophilic (94H74-124B). LGR did NOT include inflammation scored as part of the Ichthyophonus score [see below] or pigmented macrophage aggregates scores as part of the LMA score [see above].
 - A. score = 0; no granulomatous inflammation (type specimen = 94H74-1B).
 - B. score = 1; the sections had <1 focus of granulomatous inflammation per 100X field (type specimen = 94H74-7B).
 - C. score = 2; the sections had ≥1 but <3 foci of granulomatous inflammation per 100X field (type specimen = 94H74-2B, -141B).
 - D. score = 3; the sections had ≥3 foci of granulomatous inflammation per 100X field (type specimens = 94H74-37B, -127B).
- VI. EGL = eosinophilic granular leukocytes (in perivascular or pericholangial connective tissue). Note that EGLs associated with liver macrophage aggregates (LMA) and liver granulomas (LGR) were incorporated into scores for those lesions and were NOT included in this score. Here, EGLs in the connective tissue were not directly associated with any foreign material/body, but were usually associated with lymphocytes.
 - A. score = 0; ≤ 2 (and usually zero) EGLs per perivascular or pericholangial section (type specimen = 94H74-31B).
 - B. score = 1; >2 but ≤25 EGLs per perivascular or pericholangial section (type specimen = 94H74-4B).
 - C. score = 2; >25 EGLs per perivascular or pericholangial section, and EGLs extended to the margins of the surrounding parenchyma (type specimens = 94H74-96B, -152B)
 - D. score = 3; none were severe
- VII. LIP = lipidosis. A lesion in hepatocytes; excess lipid appears as clear, round, well-demarcated, cytoplasmic vacuoles (= lipid vacuoles).
 - A. score = 0; hepatocytes had no lipid vacuoles (type specimen = 94H74-1B).
 - B. score = 1; less than 33% of hepatocytes in the section had lipid vacuoles (type . specimen = 94H74-21B).
 - C. score = 2; 34-66% of hepatocytes in the section had lipid vacuoles (type specimen = 94H74-2B).
 - D. score = 3; more than 66% of hepatocytes in the section had lipid vacuoles (type

specimen = 94H74-114B).

- VIII. FPL = focal/multifocal parenchymal leukocytes. Leukocyte aggregates were usually less than 500 μ m in diameter and were composed mostly of lymphocytes and sometimes macrophages.
 - 1. score = 0; no focal parenchymal leukocytes (type specimen = 94H74-32B).
 - 2. score = 1; <1 focus of parenchymal leukocytes per 100x field (type specimen = 94H74-1B).
 - 3. score = 2; 1-2 foci of parenchymal leukocytes per 100x field (type specimen = none).
 - 4. score = 3; none were severe
- IX. ICH = hepatic Ichthyophonus.
 - A. score = 0; sections had no *Ichthyophonus* organisms (type specimen = 94H74-1B).
 - B. score = 1; *Ichthyophonus* present, but <1 per 100x field and minimal inflammation (type specimen = 94H74-166B).
 - C. score = 2; ≥1 *Ichthyophonus* per 100x field, but minimal inflammatory reaction (type specimen = 94H74-214B).
 - D. score = 3; ≥1 Ichthyophonus per 100x field, with prominent granulomatous inflammation, or ≥3 Ichthyophonus foci per 100x field, regardless of amount of inflammation (type specimens = 94H74-20B, -113B).
- X. COC = hepatic coccidian Goussia [Eimeria] clupearum. These coccidians were most common free in the parenchyma or in macrophage aggregates around bile ductules. Cysts were eosinophilic and about 18 x 12 μ m, whereas trophozoites were pale, basophilic, and about 35 μ m in diameter. Even in severe cases, inflammation associated with *E. clupearum* was minimal.
 - A. score = 0; sections had no Goussia clupearum (type specimen = 94H74-1B).
 - B. score = 1; Goussia clupearum present, but ≤ 2 foci per 100x field (type specimen = 94H74-13B).
 - C. score = 2; >2 but ≤ 6 foci of *Goussia clupearum* per 100x field (type specimen = 94H74-2B).
 - D. score = 3; >6 foci of *Goussia clupearum* per 100x field, and may be associated with inflammation (type specimen = 94H74-23B).
- XI. FN = focal necrosis. A lesion primarily of hepatocytes. Affected cells had hypereosinophilic coagulated cytoplasm, and pyknotic, karyorrhectic, or karyolytic nuclei.
 - A. score = 0; No necrotic cells in the section. (type specimen = 94H74-1B).
 - B. score = 1; total area of necrosis was $\leq 400 \ \mu m$ in diameter (type specimen = 94H74-139B).
 - C. score = 2; total area of necrosis was >400 μm but ≤1 mm in diameter (type specimen = 94H74-117B).
 - D. score = 3; total area of necrosis was >1 mm in diameter (type specimen = 94H74-207B).
- XII. SCN = single cell necrosis. A lesion of hepatocytes. Affected cells had pyknotic nuclei and condensed cytoplasm that often stained more deeply eosinophilic than normal cells. Because

of cytoplasmic collapse, individual necrotic cells were sometimes surrounded by a clear ring or halo. SCN must be differentiated from artifact. Even slightly rough handling results in cells with dark-staining cytoplasm, but nuclei were not pyknotic and cytoplasm tends to stain basophilic.

A. score = 0; No necrotic cells in the section. (type specimen = 94H74-1B).

B. score = 1; <1 necrotic cell per 400x field (type specimen = 94H74-53B near mark).

- C. score = 2; 1-2 necrotic cells per 400x field (type specimen = 94H74-118B).
- D. score = 3; >2 necrotic cells per 400x field (type specimen = 94H74-117B, -193B).

XIII. CBH = cholangitis/biliary hyperplasia. Cholangitis had lymphocytic exocytosis, with variable amounts of bile ductule hyperplasia and fibrosis.

- A. score = 0; no cholangitis or biliary hyperplasia (type specimen = 94H74-32B).
- B. score = 1; ≤ 2 foci of cholangitis or biliary hyperplasia, and foci were $\leq 400 \ \mu m$ in diameter (type specimen = 94H74-61B).
- C. score = 2; >2 foci of cholangitis or biliary hyperplasia, or foci were >400 μ m in diameter (type specimen = 94H74-112B).
- D. score = 3; none were severe.

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For statistical analysis, lesions with a score of none (0) will be used as controls. In PWS Spring samples, 240 fish will be sampled at random and used for all analyses. In addition, 20 fish will be selected because of severe lesions; these fish will be used to determine causes of morbidity, but will not be used for population prevalence calculations. The primary hypothesis to test is that fish with lesions are different from controls. The association of categorical variables (e.g., none, mild, moderate, and severe) with continuous variables (e.g., CPK values) will be determined using one-way analysis of variance (one-way ANOVA). For example, the CPK values for fish with a liver *Ichthyophonus* score of zero will be compared to livers with mild, moderate, and severe *Ichthyophonus*; when necessary, categories will be combined to ensure that each group has at least 8 fish. Category-specific means and standard errors will be calculated for each continuous variable and compared using Tukey's Studentized range method. Levene's test for equality of variances will be used to evaluate the validity of the ANOVA.

The association of selected categorical variables (e.g., *Ichthyophonus* scores versus scores for hepatic focal necrosis) will be evaluated using Chi-square methods for categorical data analysis; comparisons will be considered valid only if individual expected cell frequencies are >1 and no more than 20% of the cells have expected cell frequency <5. Odds ratios will be calculated for standard (2x2) two-way contingency tables only. To measure the strength of the linear relationships between two continuous variables, the correlation coefficient r will be calculated. For all analyses, comparisons will be considered significant when P<0.05 and highly significant when P<0.01.

Adjustments for age, gender, sampling day, and hold time will be done as necessary using multiple regression. For comparison of lesion scores and blood values by reproductive stage and site of capture, principal components analysis will be used. Similar analysis was done for the damage assessment part of fish histopathology studies funded by the Trustee Council, and results were used to separate oiled from clean sites.

C. Contracts and Other Agency Assistance

Plasma chemistry analysis, other than osmolality, will be done by Med Veterinary Lab Partners, 2231-A Commerce Ave., Concord, CA 94520 (phone: 800-432-9939; FAX: 510-689-5991); they can run 17 analytes at 25° C with only 200 μ L of plasma. The State of Alaska does not have a veterinary diagnostic laboratory, and two other laboratories either were more expensive or had equipment that could only be run at 37° C (too warm for coldwater fish enzymes). Med Veterinary Laboratory does not have a machine capable of osmolality determinations, but they will send plasma samples to UC Davis for osmolality. Other agencies will not be involved in this project.

D. Location

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Prince William Sound and Sitka Sound, Alaska. Information from this study will be of benefit to fisheries managers as they consider alternatives for managing herring fisheries.

SCHEDULE

DATES (report due on final date)	ΑCTIVITY
Fall Samples:	
Oct. 1 - Nov. 30, 1995:	Collect Fall Samples Person in charge: Gary D. Marty, UC Davis
Nov. 1 - Dec. 31, 1995:	Scale analysis (age); Person in charge: Mark Willette, ADFG, Cordova, AK
Nov. 1 - Nov. 30, 1995:	Plasma chemistries; Person in charge: Craig Ruhe, MVL, Concord, CA
Nov. 1, 1995 - Feb. 28, 1996:	Virology (includes blind passes and laboratory report) and bacteriology; Person in charge: Ted Meyers, ADFG, Juneau, AK
Nov. 1, 1995 - Feb. 28, 1996:	IgM assay; Person in charge: Ronald P. Hedrick, UC Davis, CA
Nov. 1, 1995 - Feb. 28, 1996:	Histopathology and identification of Ortholinea orientalis; Person in charge: Gary Marty, UC Davis, CA
Nov. 1 - Feb. 28, 1996:	VEN analysis and leukocyte differential counts; Person in charge: Chris Kennedy, Simon Fraser Univ
March 1- May 31, 1996:	Statistical analysis; Person in charge: Thomas Farver, UC Davis, CA

A. Measurable Project Tasks for FY96

Spring Samples

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DATES (report due on final date)	ACTIVITY
March 1 - April 30, 1996:	Collect Spring Samples Person in charge: Gary D. Marty, UC Davis
April 1996	Visit native harvesters in Tatitlek and lead an open discussion for fishers in Cordova (Person in charge: Gary D. Marty, UC Davis)
April - July 31, 1996:	Scale analysis (age); Person in charge: Mark Willette, ADFG, Cordova, AK
April - May 31, 1996:	Plasma chemistries; Person in charge: Craig Ruhe, MVL, Concord, CA
April - Sept. 30, 1996:	Virology (includes blind passes and laboratory report) and bacteriology; Person in charge: Ted Meyers, ADFG, Juneau, AK
April - Sept 30, 1996:	VEN analysis, leukocyte differential counts, and CPK isozyme analysis; Person in charge: Christopher Kennedy, SF Univ., BC
April - Sept 30, 1996:	IgM assay; Person in charge: Ronald P. Hedrick, UC Davis, CA
April - Sept 30, 1996:	Histopathology and identification of Ortholinea orientalis; Person in charge: Gary Marty, UC Davis, CA
Oct. 1996 - Jan. 10, 1997:	Statistical analysis; Person in charge: Thomas Farver, UC Davis, CA
Jan. 11, 1997 -April 15, 1997:	Annual report writing Person in charge: Gary Marty, UC Davis, CA
Nov. 1996 - indefinite:	Opportunities for public comment

B. Project Milestones and Endpoints

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1. **Objective:** Determine which pathogens cause lesions and determine the relation among Viral Hemorrhagic Septicemia virus (VHSV), *Ichthyophonus*, macroscopic and microscopic lesions, plasma chemistries, and immune status.

When objective will be met: the annual report, due April 15, 1997, will provide information progressing towards this objective, but the most complete information will not be available until after the multi-year study is completed and the final synthesis report is submitted April 15, 1999.

Objective: Determine the role of reproductive stage on the general health of herring. Are lesions, *Ichthyophonus*, and VHSV more severe during a given reproductive stage? When objective will be met: the annual report, due April 15, 1997, will provide the first information progressing towards this objective, but the most complete information will not be available until after the multi-year study is completed and the final synthesis report is

submitted April 15, 1999.

3. Objective: Investigate the impact of disease on population size and structure of herring. Are fish of a particular year class more likely to be diseased than fish of other year classes? Does a history of previous oil exposure correlate with prevalence and severity of disease? When objective will be met: based on study from 1994-1996, the annual report, due April 15, 1997, will provide information progressing towards this objective, but the best information will not be available until after the multi-year study is completed and the final synthesis report is submitted April 15, 1999.

C. Project Reports

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Annual reports will be submitted to the Chief Scientist on April 15, 1997 (FY96) and April 15, 1998 (FY97). A final report will be submitted after field work is completed in FY98: April 15, 1999. After publication of results from study in 1994 (94320S), publication of additional results will be most useful at the end of the multiyear study. The Journal of Aquatic Organisms is probably most appropriate for these data.

COORDINATION AND INTEGRATION OF RESTORATION EFFORT

This project was included in 95320, the Prince William Sound System Project, because major fluctuations in the population size of PWS herring may have repercussions on many other injured resources in PWS. Reduction of the herring population to 10% of what might have been expected undoubtedly has a significant impact on those resources for which herring are a major dietary component. Understanding the population dynamics of herring in PWS will be addressed by the coordinating information on diseases (96320-S, this subproject) with many other projects and subprojects of 96320. Herring spawn deposition (ADFG project) and reproductive impairment (96074) address several aspects of recruitment. Herring genetics (95165) will determine whether there is more than one stock in PWS. Zooplankton in the ecosystem (96320-H) will determine what is available for herring to eat. Forage fish (96163-I) and avian predators (96320-Q) examine the relationship between herring and those species that feed on them. Most other parts of 96320 also provide some information on the factors which affect herring populations as well as other parts of the Prince William Sound ecosystem. Synthesizing the results of all of these projects and subprojects should document the reasons for major changes in herring population size, structure, and health status.

Specific interactions are as follows: 1) fish captured at the same time as disease samples will be available for reproductive impairment (96074), genetics (96165), and age-weight-length studies conducted under normal ADFG management or research studies; 2) the Fall hydroacoustic surveys (96320N) and natal habitats (96166) will be used to locate and capture fish for Fall disease samples in PWS; and 3) the University of California, Davis (Dr. Gary D. Marty) is under contract with NOAA to examine tissues from herring adults exposed to hydrocarbons at the Auke Bay Laboratory (95074); examination by the same pathologist will ensure comparability of field and laboratory studies. Normal agency management overlapping with this project is limited to age-weight-length studies of prespawning and spawning fish, and ADFG is supporting that part of the project with non-Trustee funds. Also, ADFG is supporting all salaries of personnel doing virology for the project at the Juneau Fish Pathology Laboratory; this is consistent with the goal of ADFG to monitor Alaskan fish for disease. The purpose of normal ADFG study is to follow normal population trends. But the decline in PWS herring numbers since the spill is unprecedented in the history of Alaskan herring fisheries. Therefore, the detailed study we propose has never been required for management decisions. If components of this study prove cost effective (e.g., examining fish for external lesions), we will recommend their incorporation into protocols for normal population monitoring. Attempts to obtain matching funds from non-Trustee Council sources have not been made.

ENVIRONMENTAL COMPLIANCE

The National Oceanic and Atmospheric Administration (NOAA) is the lead federal agency for National Environmental Policy Act (NEPA) compliance for this project. This project has previously been granted categorical exclusion because it is essentially a laboratory study without environmental consequences. Samples will be collected by the ADFG personnel under the authority of a scientific collector's permit issued by ADFG.

PERSONNEL

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Project Leader (Field Component):

Gary D. Marty, DVM, and Diplomate, American College of Veterinary Pathologists, will be responsible for design of pathology studies, on-site necropsy evaluation, reading histologic preparations, and final report writing. Dr. Marty has the required fisheries background (B.S. and M.S. in fisheries biology) to integrate the many parts of this study, and he performed these duties on project 94320S and 95320S.

Other Key Personnel (Field Component):

Corrine R. Davis, DVM, will do on-site necropsy evaluations, read histologic preparations, and develop an immunoglobulin assay. Dr. Davis preformed necropsies on Pacific herring in 1994 and 1995 (94320S and 95320S), and she was the primary histopathologist for analyzing gonads from those fish.

Thomas B. Farver, Ph.D., is professor of biostatistics and has done extensive consulting on problems of statistical epidemiology, including project 94320S and 95320S. He will oversee statistical analysis.

Ronald P. Hedrick, Ph.D., is a professor and Chief of the Aquatic Medicine Service, Veterinary Medical Teaching Hospital, University of California, Davis. Dr. Hedrick is certified as a Fish

Pathologist by the Fish Health Section of the American Fisheries Society and has extensive experience with infectious diseases and immunology of fish. He is overseeing development of the IgM assay (95320S) and will oversee immunoglobulin analysis as part of this project.

David E. Hinton, Ph.D., is professor and director of the Aquatic Toxicology Laboratory at the University of California, Davis. Dr. Hinton has extensive experience in fish toxicology and histopathology. He will be in charge of project administration at the University of California, Davis.

Theodore R. Meyers, Ph.D., is certified as a Fish Pathologist by the Fish Health Section of the American Fisheries Society. Dr. Meyers has been Principal Pathologist for the AK Dept. of Fish and Game since 1985. Dr. Meyers and the laboratories he supervises have been involved in the detection and diagnosis of VHSV in Alaskan fisheries since 1990, detecting the virus in cod and herring from PWS and in herring from other parts of Alaska. Dr. Meyers is the designated principal investigator for virology and bacteriology on this project.

Relevant Publications:

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Section II

Laboratory challenge of Pacific herring with and without stressors

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J.R. Winton National Biological Service 7500 Sandpoint Way NE Seattle, WA 98115 (206) 526-6587 PROJECT DESIGN: II. Laboratory challenge with and without stressors.

A. Objectives

<u>FY 96</u>

- 1. Establish SPF herring in the laboratory for use in definitive disease studies on VHSV and *Ichthyophonus hoferi*
- 2. Fulfil Koch's Postulates for VHSV in SPF Pacific herring
- 3. Fulfil Koch's Postulates for *I. hoferi* in SPF Pacific herring
- 4. Establish an SPF model system for studying VHSV and *I. hoferi*.

<u>FY 96 - 97</u>

5. Describe the effects of physical and chemical stressors on Pacific herring in the absence of disease organisms.

<u>FY 97 - 98</u>

- 6. Describe the effects of physical and chemical stressors on the course of disease produced by VHSV and *I. hoferi*
- 7. Describe the immune response and blood chemical changes associated with infection by VHSV and *I. hoferi.*
- 8. Describe how exposure to chemical and physical stressors can affect the course of disease produced by VHSV and *I. hoferi*.
- 9. Describe the course and outcome of multiple infections in Pacific herring.

B. Methods

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o Quarantine Facility (In place and available at NBS: see attached letter)

Virus-free water source

The majority of the herring rearing and exposure studies will be carried out at the Marrowstone Island Field Station of the National Biological Survey. This facility is located on Marrowstone Island on Admiralty Inlet (Puget Sound, WA). Seawater will be pumped from 60 ft below the surface of Admiralty Inlet through a sand filter and U.V. sterilization system before being used in the study. This is an area of fast flowing water with no herring spawning activity within several miles. The treated water will be cultured for bacterial, fungal and viral contaminants using standard microbiological techniques and cell cultures susceptible to VHSV. Monitoring of the water will take piace prior to the study and monthly throughout the study period.

Flow-through sterile seawater

During incubation the seawater will be constantly monitored for dissolved oxygen and pH, and adjusted if any change from optimum conditions occur. At the time of hatching the water will be replaced at the rate of two full exchanges per day to remove perivitelline fluid, chorion husks and other proteinaceous materials which might act as microbial growth media. This low flow rate is adequate to sustain the newly hatched larvae as well as remove any toxic metabolites, but not so high that the animals are damaged. As the larvae grow, the water flow will be gradually increased to accommodate the greater depuration of metabolites from the larger fish.

Flow-through natural seawater:

A parallel set of two tanks will be used to monitor the effectiveness of the seawater sterilization process. The embryos and larvae will be treated as described above, except that the tanks will receive raw unfiltered seawater. This should give an indication of the effectiveness of filtered incubation water on the natural transmission of pathogens to larval herring when the water is not associated with heavy herring use.

Physical isolation of control and treated fish

During the course of the studies, SPF herring will be separated from test fish by both physical barriers within the wet lab as well as separate water supplies. All equipment used to handle fish will be maintained separately for each tank and stored in disinfectant when not in use. Subsamples of fish will be taken monthly and examined for VHSV by infectivity cell culture assays, and tissues taken for histopathologic examination for *lchthyophonus* as well as other potential pathogens.

Depurated effluent

Water used for pathogen and toxin exposure will be chemically disinfected before leaving the Marrowstone facility to ensure that pathogens are not escaping the facility and entering the natural marine waters of Admiralty Inlet. After the water is treated it enters a settling pond before draining into Admiralty Inlet.

Task 1: Fish (FY 95 thru 98)

Task 1.1: Obtaining & hatching herring eggs

Initially, herring eggs will be obtained from Prince William Sound in conjunction with ADF&G Spawn Deposition Surveys. Herring for the SPF study will be produced from artificially spawned eggs incubated in sterile seawater as described by Kocan et al (1995). Spawning adults will be captured by net and their surface sterilized with iodophore and alcohol. Eggs will be removed from the females and broadcast onto an artificial substrate, fertilized with milt from surface-sterilized males and allowed to incubate in sterile seawater until they hatch. Following fertilization, the eggs will be transported by commercial air carrier to the University of Washington and the Marrowstone Island Field Station as previously described by Kocan et al (1995). A contingency or back-up system will consist of eggs obtained from Puget Sound herring and incubated in parallel with those obtained from Prince William Sound. This will ensure that if problems arise with one set of embryos that the project will not be jeopardized. If both egg lots survive, then comparative cata between the two populations will be generated.

Task 1.2: Rearing Herring Larvae to adults

Newly hatched Pacific herring larvae will be reared in flow-through seawater systems with constant aeration in a system similar to that described by Talbot and Johnson (1972), and used by various Aquariums for the rearing of larval fish. Water temperature, pH and oxygen will be monitored daily. The water will be beriodically conditioned with algal paste (as needed) according to the protocol described by Marliave and Whyte (Vancouver,B.C. Aquarium), and the larvae fed brine shrimp hatched in sterile seawater and supplemented with omega-3 fatty acids. Tetramin@ baby-fish food will be used as a supplement feed. Once the larvae reach 2 cm they will be fed frozen adult brine shrimp and live lab-reared

daphnia for the duration of the studies. Larvae should grow at about 10 mm per month, and have been shown to survive in captivity for at least 2 years (Talbot and Johnson 1972).

Pilot larval rearing studies will be conducted on 0-age class herring larvae captured by tow net just off the shore at the Marrowstone Island Field Station. This will be accomplished by use of a charter vessel and the NBS skiff stationed at Marrowstone Island. The larvae will be used to establish the protocols necessary to rear SPF larvae from the artificial spawn described above as well as to establish methodology for handling, dosing, sampling and evaluating the health of laboratory-reared herring. If these fish prove to be free of either VHSV or *I. hoferi*, they will be used in some of the stress-related experiments.

Puget Sound herring are sexually mature and actively spawn at 2 years, while Prince William Sound fish first spawn at 4 years-old. If this is a genetic rather than geographic difference, SPF spawning herring could be available in 2 years by using Puget Sound fish for reproductive (spawning fish challenge) studies.

Uniform size and age class

Fish will be segregated by age class throughout the course of these studies. Each age class will also be graded and further segregated by size in order to minimize variability among treatment groups and controls. Fish from different sources (eg. PWS and PS) will not be mixed, with the possible exception of studies intended to show contact transmission of pathogens in the laboratory.

Task 2: Verification of SPF for VHSV and Ichthyophonus

Once larvae begin feeding, and prior to the initiation of experiments, subsamples of larvae will be collected and screened histopathologically and by *in vitro* culture to verify that the fish are free of VHSV and *lchthyophonus*. (Fish Health Blue Book of the American Fisheries Society, Thoesen, 1994). This screening will continue for all stocks of natural or artificially spawned fish throughout the course of these studies.

Task 2.1: Histopathology

For histopathological examination, 25 randomly selected fish will be sampled from the population. Tissues that are particularly sensitive to one or both pathogens (i.e. kidney, liver, spleen, heart) will be analyzed. Additional tissues (gill, muscle, gonad, brain, g.i. tract, pancreas) will be collected, preserved and stored for later examination should that be deemed necessary. The fish will be anesthetized in MS-222, sacrificed by severing the spinal cord, and examined for the presence of grossly visible lesions. The target tissues, as well as any obvious lesions. will be removed during necropsy, preserved in 10% neutral buffered formalin, embedded in paraffin, sectioned on a rotary microtome (5 μ m) and stained with hematoxylin and eosin (Luna, 1974). Tissues will be examined by light microscopy. For the sake of consistency, the results of the laboratory study will be reported using the terminology and scoring system developed for the Component 1 field study (Marty et al., 1994).

Task 2.2: In vitro culture of Ichthyophonus

Kidney, liver, and heart tissue will be aseptically removed from 25 randomly selected fish. The tissue will be cut into small pieces ($\geq 2 \text{ mm}^3$), immersed briefly (1-2 sec) in ethanol and placed in tissue culture flasks containing Eagles minimal essential medium (MEM) supplemented with 10% fetal bovine serum. 3.5% NaCl, and 100 units of penicillin/streptomycin. The cultures will be incubated at 15-20 C and examined for the production of hyphae and endospores.

Some cultures will be maintained in serial passage for use during the remainder of the study. Endospores and possible resting spores will be harvested, quantitated and used for experimental inoculations.

Task 2.3: In vitro culture of VHSV

Homogenates of kidney and spleen tissue collected from 25 randomly selected fish will be filtered through a 0.45 μ m filter and cultured on the EPC cell line at 15 C (pH 7.4-7.6). The cells will be examined for evidence of cytopathic effect. Cultures will be maintained for 14-21 days, and may be blind passaged after 14 days. Should cytopathic effects be observed, the identity of the causative agent will be determined by serum neutralization assays.

Task 3: Challenge without stressors (FY 96)

Koch's Postulates. A series of of experimental criteria first applied by Robert Koch, are necessary to establish the causal relationship between a specific microorganism and a specific disease. These criteria include:

- 1. The microorganism must be present in every case of the disease.
- 2. The microorganism must be isolated from the diseased host and grown in pure culture.
- 3. The specific disease must be reproduced when a pure culture of the microorganism is inoculated into a healthy susceptible host.
- 4. The microorganism must be recoverable from the experimentally infected host.

The first two criteria will be or have already been met by isolating both VHSV and *I. hoferi* from Prince William Sound herring and establishing them in pure culture. Criteria 3 and 4 are described below.

Task 3.1: Challenge herring with VHSV.

The North American strain of VHSV obtained from adult herring in Puget Sound. Washington in 1994 will be used in this study. This virus is identical to that isolated from Prince William Sound herring. The virus will be grown in the epithelioma papullosum cyprini (EPC) cell line to titers of approximately 10⁷ plaque-forming units per ml. Replicate groups of 30 herring will be challenged by waterborne exposure to 10², 10⁴ or 10⁶ PFU/ml seawater in a static bath for 1 hr. Exposed fish and unexposed controls will be held for 21 days and examined daily for mortality or signs of disease. Additional replicate groups of 30 herring will be challenged by intraperitoneal injection of 10², 10⁴ or 10⁶ PFU of VHSV per fish. Fish will be observed daily as above. After 21 days, virus will be re-isolated and new SPF fish will be exposed to complete Koch's Postulates. These will be treated as in the original group of infected fish.

Blood will be collected from a subsample of infected herring after 21 days and tested for the presence of antibodies to VHSV by virus neutralization, Ouchterlony gel diffusion or countercurrent electrophoresis. This information will be use as a baseline for studies carried out in FY 96-97 on "Challenge With Stressors".

Task 3.2: Challenge herring with Ichthyophonus.

I. hoferi isolated from Prince William Sound herring tissues will be grown in minimal essential medium plus 10% FBS (MEM-10) and used for initiating infections in experimental fish. Graded doses of *in vitro* derived spores will be used to orally infect replicate groups of 30 herring. Fish will be subsampled (10 ea) at 14 days and the remainder maintained in flowing sterile seawater for a total of 30 days post infection. Mortality and morbidity will be recorded at this time and the fish sacrificed for histopathology and re-isolation of the organism.

Organisms isolated these fish will be used to reinfect new fish and complete Koch's Postulates. Based on the available literature (Sinderman and Chenoweth 1993), it may be possible to obtain *lchthyophonus*-free fish by capturing 0-age fish and maintaining them in pathogen-free seawater. This would remove some of the pressure on production of enough SPF fish during FY 95.

Blood will be collected from a subsample of infected herring after 30 days and tested for the presence of antibodies to *I. hoferi* by ouchterlony gel diffusion and counter current electrophoresis. This information will be use as a paseline for studies carried out in FY 96-97 on "Challenge With Stressors".

Task 3.3 - Assay experimental fish for VHSV and Ichthyophonus.

Moribund and diseased fish will be removed from rearing tanks daily. Samples of diseased fish will be collected and assayed for levels of VHSV and *Ichthyophonus* by standard methods. Additional material will be collected from diseased fish and processed for histopathological examination. At the end of the challenge period, samples will be collected from diseased fish for virology and histology. The virus and *Ichthyophonus* isolated from diseased fish will be identified using standard methods.

Task 3.4: Statistical Analyses

Task 3.4.1 Analyses for larval rearing will consist of:

- % hatch
- % larval survival to feeding
- % larval survival by month
- larval growth rate by month

Task 3.4.2 Analyses for effect of VHSV infection:

- infection rate (% infected fish)
- virus titer per fish
- overt disease (eg. visible lesions)
- mortality (control vs infected)
- comparison of water-borne vs inoculation infections

Task 3.4.3 Analyses for effect of *Ichthyophonus* infection:

- infection rate
- infection intensity
- overt disease (eg. visible and microscopic lesions)
- mortality (control vs infected)
- comparison of water-borne vs feeding infections

Task 4: Density as a stressor (FY 96 -> 98)

Task 4.1: density + pathogens

Ideally, it would be desirable to use SPF fish that have been experimentally infected with a known dose of pathogens for the density dependant disease studies. Once density dependent disease conditions are established uncer controlled conditions, then wild-caught fish could be tested to determine whether they respond similarly under identical conditions. Experimental fish will be evaluated for mortality, gross lesions, microscopic lesions, VHSV or *I. hoferi* infection and behavioral changes. Blood samples will be collected and evaluated by Dr. Chris Kennedy (Simon Fraser) for biochemical changes. Any observed lesions will be compared with those seen in PWS in wild herring.

If laboratory reared fish are of suitable size and age prior to the end of year-1, they will be used for the initial study. Otherwise, juvenile wild fish (>5 gm) will be captured by tow net from Puget Sound and transported to the Marrowstone Island Field Station. Fish will be acclimated in a 1,000 gal tanks for two weeks, then transferred to 200 gal tanks for density dependant disease studies. Half of the fish will be untreated and half will be inoculated with a known dose of VHSV or *lchthyophonus* in order to have a control (or reference) test population and one with a known infection rate. Initially, fish densities will be 25, 50, 100 and 250 fish per 200 gal, with two replicate tanks per density. Densities will be modified later if warranted. Flow rate, temperature and feeding schedule will be constant for all tanks. Fish will be observed several times per day and moribund or cead fish removed for 30 days. At this time fish will be sacrificed as described above, blood samples taken for evaluation of neutralizing antibodies and tissues prepared for virus isolation or histopathology.

We will use concentrations of the North American strain of VHSV and of *lchthyophonus hoferi* which are shown in Task 3.1 and 3.2 to produce a low to moderate level (\leq 20%) of mortality by intraperitoneal injection into fish held at a density of 30 fish per tank. Replicate groups of herring will be placed into flowing seawater aquaria at 4-5 densities for challenge by VHSV or *lchthyophonus* (Fig. 1).

Experiments on density-stressed infected fish will begin in FY 96 following the establishment of Koch's Postulates. These studies are based on the assumption that both organisms are capable of producing disease in Pacific herring uncer the conditions tested. If it turns out that one of the organisms is not a pathogen in herring, then testing will proceed with only one organism.

Fish / tank



chthyophonus-infected fish

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Figure 1. Scheme for studying density-dependant stress on control and infected Pacific herring.

Experimental conditions:

Flow-rate	≥ 50 gph
Tanks:	200 gal
Water:	.Sterile seawater
Organisms:	. VHSV & I. hoferi
Controls:	Uninfected herring
Temperature	ambient (6º - 10º C)
pH	. ambient (8 - 9)
salinity	. ambient (25 ppt - 28 ppt)
replicates	3 / density
	-

Expected results from the Density (without stressor studies): Effect of density on SPF herring survival, growth and health Effect of density on SPF herring infected with a single pathogen Effect of density on wild herring infected with a known pathogen superimposed on their natural pathogens.

Task 4.2: pathogens with stressors (FY 96, 97)

Studies on challenge infections with stressors will begin in FY96 following the completion of the density dependent disease studies. Once optimum densities for fish survival in the absence of pathogens have been determined, (eg. Task 4.1-controls) studies will commence on the effects of stressors on pathogen-infected fish. Experimental fish will be evaluated for mortality, gross lesions, microscopic lesions, VHSV or *I. hoferi* infection and behavioral changes. Blood samples will be collected and analyzed by Dr. Chris Kennedy (Simon Fraser) for biochemical changes. Any observed lesions will be compared with those seen in wild PWS herring.

<u>Task 4.2.1: Chemical stress of pathogen-infected fish</u> (FY 97, 98) Replicate groups of 25 herring will be placed into flowing seawater tanks at optimum density for infection by intraperitoneal injection of three doses of the North American strain of VHSV or *lchthyophonus hoferi* which were shown to produce a low to moderate level ($\leq 20\%$) of mortality in herring held at a density of 30 fish per aquarium at ambient temperature seawater in Puget Sound (approximately 8-9°C). Chemical stressors will be added to the system 5 days post-infection by means of a metered pump. It has been demonstrated that crude oil introduced to a population of naturally infected herring will cause an increase in infection rate (Exhibit 8; Carls & Meyers). Consequently, components of crude oil known to have immunosuppressive activity will be used for the chemical stress of pathogen-infected fish. Tests will include but are not restricted to whole Prudhoe Bay crude oil and its components

Chemical stressor concentrations will vary with the solubility of the compound(s) being tested and the established toxic levels reported in the literature. Both PAH and alkanes have been shown to be immunosuppressive in vertebrates, but have not been investigated in fish. This experiment will define their effect(s) on the immune system and ultimate susceptibility to the pathogens being tested.

Serum will be collected from pre- and post-exposed fish and evaluated for changes in neutralizing antibodies to VHSV and *I. hoferi.*

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Controls for chemical stessors will consist of pathogen-free fish exposed to the same concentrations of petroleum as the infected fish. Controls will be run in parallel with the test fish and be of the same age, size and origin (Figure 2).

Analytical evaluation: Water from each test tank will be collected in acid washed glass vessels and analyzed for petroleum hydrocarbons and individual hydrocarbon groups. Following exposure, a subsample of fish will also be collected for tissue analysis of hydrocarbon content. The effect of oil exposure on previous infections by VHSV and *I. hoferi* will be determined.

Task 4.2.2: Pathogen challenge of chemically stressed fish (FY 97, 98) In this study herring will first be chemically stressed by exposure to crude oil, then infected with a known sub-lethal dose of the two pathogens.

Fish will be set up in tanks supplied with sterile seawater at 30 fish per tank and exposed to three concentrations of petroleum hydrocarbons at concentrations which do not produce overt signs of distress. The fish will then be exposed to VHSV or *lchthyophonus* 5 days later at a dose which produces $\leq 20\%$ mortality. The fish will be held for 30 days and observed for mortality and assayed for virus or *lchthyophonus*. Concentrations of the oil components will be calculated based on the data reported by Carls and Meyers (Exhibit 8) and the scientific literature. These would begin at the proportion of each component expected to be present in 300 µg/L (300 ppb) whole crude oil, and include 3 ten-fold dilutions. Actual concentrations will be determined by chemical analysis of water collected during the exposure period.

Serum will be collected from pre- and post-exposed fish and evaluated for changes in neutralizing antibodies to VHSV and *I. hoferi*.

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Controls will consist of tanks receiving no pathogen challenge (hydrocarbon only) and tanks receiving raw (non-sterile) seawater. The general design of this study (without replicates) is presented in Figure 3.



Figure 2. Oil stress of pathogen-infected fish

Scheme for exposing Pacific herring to Prudho Bay crude oil with and without pathogens. Multiple doses of hydrocarbon will be used with multiple infective doses of pathogen.




perimental conditions:
Flow-rate ≥ 50 gph
Temperature ambient (8° - 10° C)
pH ambient (8 - 9)
salinityambient (25 ppt - 28 ppt)
replicates
HC concentrations 3
Pathogen dose \leq 20% mortality in non-stressed fish

Task 5: Co-infections (FY 97, 98)

A non-lethal dose level for both pathogens will be established in Component 2. Once this data is available on pathogen doses producing $\leq 20\%$ mortality, concurrent infections will be produced by infecting fish with both organisms simultaneously and in sequence. Specific conditions related to the implementation of this task have not been worked out at this time, but once preliminary data on dose related mortality and disease is generated a more comprehensive study plan can be designed. The basic exposure scheme will be modeled on that described under Component 2.

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C. Contracts and other agency assistance

No outside contracts. Assistance by DOI - NBS will continue throughout the project period.

D. Location

Field collections will be made in Prince William Sound, Alaska in conjunction with ongoing ADF&G activities or under contract with local fishermen during the normal fishing season(s). As much of the needed material as possible will be collected on-site in Prince William Sound, while the remainder will be obtained from Puget Sound by the University of Washington School of Fisheries and Friday Harbor Labs, the National Biological Survey, and the Marrowstone Island Field Station (Puget Sound, WA). These laboratories have the necessary containment facilities for working with VHS, *Ichthyophonus* and other pathogens, and the seawater systems for carrying out the *in vivo* VHS-free portions of the study. Collection of herring eggs and 0-age herring in Puget Sound will be done under contract to the Mr. Charles Eaton (R.V. Kittiwake) under a scientific collector's permit issued to R.M. Kocan by the Washington Dept. of Fisheries. Blood samples collected from experimental fish at the quarantine facility will be transported to Simon Fraser University for final analyses by Dr. Chris Kennedy.

SCHEDULE

A. Measurable Project Tasks for FY 96

May-Dec. 1995	Culture herring larvae and determine their SPF status for future work. Collect data on growth, survival, disease susceptibility, etc. Improve husbandry techniques where possible.
Oct '95- Dec. '95	When larvae are large enough, begin viral and fungal exposures to determine susceptibility.

Continue or begin infectivity studies with VHSV and I. hoferi. Jan - June 1996 Determine LC50 for both organisms, minimal infective dose, survival rate, lesions associated with infection by each organism. and recovery or carrier rate. April - May 1996 Begin new year of SPF fish from eggs for future studies. March-June 1996 Re-isolate organisms and verify that monoxenic infections were produced in order to fulfill Koch's Postulates. Begin blood chemistry on infected fish and physiological studies. Collect 0-age herring for stress exposures technique June-Sept. 1996 development. May-Dec. 1996 Analyze data from infectivity - disease - survival studies and begin studies on stress effects on infected fish. Density effects, oil effects, etc. Begin immune suppression studies on experimental fish for comparison with data from wild fish (PWS).

B. Project Milestones and Endpoints

Oct. - Dec. '95 March '96 May '96 March - Sept. '96 Oct. - Dec. '96 Jan. - March. '97 March - Sept. '97 Oct. - Dec. '97 Jan. - June '98 June - Sept. '98 Aug. - Dec. '98

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Establish initial SPF herring for later studies Establish SPF model for studying VHSV & *I. hoferi* Fulfil Koch's Postulates for VHSV in model system Fulfil Koch's Postulates for *I. hoferi* in model system Describe effects of chemical stressors on herring Describe effects of physical stressors on herring Describe effects of stressors & pathogens on herring Collect and analyze blood samples from experimental fish Evaluate immune response in chemically stressed fish Evaluate immune response in physically stressed fish Evaluate immune response in fish with multiple infections

C. Project Reports

Preparation of manuscripts for peer reviewed publications will begin as studies are completed and sufficient data is available. Pre-prints and reprints of these will be forwarded to the Trustee Council and Chief Scientist as they are received.

Dec. '95	Progress report for FY 95
Dec. '96	Progress report for FY 96
Dec. '97	Progress report for FY 97
Dec. '98	Final report for FY 95 through FY 98

COORDINATION AND INTEGRATION OF RESTORATION EFFORT

Alaska Dept. of Fish and Game will contribute to this project by supplying a working platform for artificial spawning of herring in PWS, assistance in capturing and spawning the fish and transportation of embryos between the collection site, Cordova and the airport. The National Biological Service (NBS) will contribute Dr. Winton's salary as well as space and equipment. Statistical consultation (project design / data analyses) will be obtained through the UW Center for Quantitative Science. Computer services (data entry, data analysis, word processing) will be provided by SOF and NBS. Histological processing of tissue samples will be done through the UW Dept. of Pathology and histopathological evaluation of tissues from experimental infections and challenges will be conducted at SOF. Cell culture, virology and molecular biology facilities will be provided by NBS. Filtered seawater facilities for contaminant exposure studies are available at the Marrowstone Island Field Station (NBS), as is sterile (VHSV-free) seawater for *in vivo* virus studies. Filtered seawater facilities are also available at Friday Harbor Laboratories (UW).

ENVIRONMENTAL COMPLIANCE

The National Oceanic and Atmospheric Administration (NOAA) is the lead federal agency for National Environmental Policy Act (NEPA) compliance for this project. This project has been granted categorical exclusion because it is essentially a laboratory study without environmental consequences. Fish infected with pathogens will be housed in an approved government facility designed and approved for pathogen studies and all effluents will be decontaminated. Samples will be collected by ADF&G personnel under authority of a scientific collector's permit issued by the ADF&G. Permits needed for work in the State of Washington are granted by Washington Dept. of Fish & Game to the Univ. of Washington (R.M. Kocan, P.I.). Collection of herring eggs and 0-age herring in Puget Sound will be done under contract to the Mr. Charles Eaton (R.V. Kittiwake) under a scientific collector's permit issued to R.M. Kocan by the Washington Dept. of Fisheries. Animal Care Committee approval of the study has been granted at the Univ. of Washington. Studies conducted by Simon Fraser University (SFU) will be coordinated with both the Field and Laboratory components of this project. Interactions will involve S.F.U. evaluation of blood chemistry from PWS fish and laboratory infected fish. Some studies will be conducted by SFU personnel at the Marrowstone Island facility because of its isolation and containment features. Data will be continually reviewed and synthesized by all three groups (U.C. Davis, U of W and SFU).

PERSONNEL

C. as

Richard M. Kocan, Ph.D.

Over the past ten years I have nad extensive experience with petroleum induced toxicity in aquatic organisms. Since 1990 I have been actively involved in the Exxon Valdez oil spill studies in Prince William Sound and have served as a peer reviewer for salmonids, rockfish, shellfish and herring during the Damage Assessment and Restoration phases of the program, as well as an expert witness for the State of Alaska and NOAA. From 1990 to 1993 I worked with the Alaska Department of Fish & Game in Cordova as a subcontractor on Herring Sublethal Effects (project #11), studying toxic and genetic effects of oil on herring embryos and larvae. Prior to working in Prince William Sound, I worked on oil related problems in Puget Sound associated with near-shore damage and evaluation of oil originating from several shore-based oil operations. These include the Cherry Point shoreline where Texaco, BP Petroleum, ARCO and Intalco Aluminum Co. are located, as well as Fidalgo Bay, where Texaco has a transfer dock and refinery. These studies were originated by the State of Washington and the Lummi and Klallam Indian Tribes and were funded by both the State and the various industries.

In 1985, prior to working on oil related problems in Puget Sound. I spent several months studying with Drs. Westernhagen and Rosenthal at the Biologische Anstalt Helgoland in

Germany. There I worked on cod, flatfish flounder, herring and turbot embryos and larvae exposed to petroleum contaminated seasurface microlayer in the Baltic Sea and North Sea.

Over the years I have developed techniques which allow for "on site" exposure of animals in contaminated marine waters as well as aboratory evaluation of sediments for toxicity to marine vertebrates and invertebrates. I have access to flowing seawater research facilities at the University of Washington, the National Biological Survey field station on Marrowstone Island, Washington and have discussed the use of the Prince William Sound Science Center facilities in Cordova with Dr. Gary Thomas.

James R. Winton, PhD

Dr. James Winton received a PhD in Microbiology from Oregon State University in 1981 where he studied fish diseases under the direction of Dr. John Fryer. After graduation, he remained on the faculty and directed the fish health research activities at the Hatfield Marine Science Center in Newport, Oregon. During that period, he had faculty appointments in the Departments of Microbiology, General Science, and Fisheries and Wildlife. While at the Marine Science Center, he did research on fish diseases, helped establish a diagnostic and certification service for private aquaculturists, and participated in international programs. His research interests include infectious diseases of fish, poikilothermic cell and tissue culture, and virus diseases of fish and shellfish.

In 1986 Dr. Winton moved to the US Fish and Wildlife Service, National Fisheries Research Center in Seattle where he serves as the leader of a fish health research team consisting of more than 20 researchers, technicians, graduate students and visiting scientists working on infectious diseases of Pacific saimon and trout. The Center is now part of the Department of Interior, National Biological Survey. As an affiliate professor at the University of Washington, he helps direct the research of graduate students working at the Center and provides lectures on fish viruses. In the past six years, he has taught the virology portion of two week Fish Disease Course at the Hatfield Marine Science Center and part of the Fish Health Long Course at the National Fisheries Center at Leetown, W. Va. Dr. Winton served as co-editor of the Fish Health Section Newsletter from 1984-1989 and is currently the subject editor for fish pathology for the Journal of Applied Ichthyology and an editorial advisor for Diseases of Aquatic Organisms. He is a Certified Fish Pathologist and a member of numerous scientific and honorary societies. He also serves on the International Committee on Taxonomy of Viruses, the American Type Culture Collection Advisory Committee, and the Fish Disease Commission of the Office of International Epizootics in Paris, France.

During the last 5 years, Dr. Winton has worked extensively with VHSV including his role in identifying the first isolates of VHSV from North America. Since that initial discovery, workers in his laboratory have characterized the North American isolates serologically and biochemically, developed DNA probes for detecting and differentiating isolates of VHSV, and conducted challenge experiments of eight species of salmonid fish showing the North American strain of the virus was different than the European type. Recently, he has assisted in the characterization of the isolates of VHSV from cod and herring in Alaska and has worked closely with Dr. Ted Meyers and the fish pathology staff of ADF&G with whom he shares authorship on several relevant papers. He is an author on more than 70 scientific publications, those dealing with VHSV are listed below.

Marsha L. Landolt, PhD

Dr. Marsha Landolt received a PhD in Pathology from George Washington University in 1975. From 1970 until 1974 she was employed as a histopathologist by the Eastern Fish Disease Laboratory in Leetown, West Virginia (US Fish and Wildlife Service). She conducted research on a variety of infectious diseases affecting trout and salmon and was an instructor in the Laboratory's Long and Short Courses on Fish Disease. From 1974 to 1975 she served a Pathology Clerkship at the National Zoological Park in Washington, D.C. In that capacity she performed post mortem examinations of all collection animals that died and conducted comparative pathological analyses on amphibian, reptilian, avian and mammalian tissues.

In 1975, Dr. Landolt became a faculty member at the University of Washington School of Fisheries. She attained the rank of Professor in 1986. As a faculty member, Dr. Landolt has taught undergraduate and graduate level courses in fish and invertebrate pathology, and she has supervised the thesis research of more than 20 graduate students. Her research at the University of Washington has focused on non-infectious as well as infectious diseases of fishes and has been supported by the National Institute of Environmental Health Sciences. Sea Grant, NOAA's Saltonstall-Kennedy Program and the US Department of Agriculture. In collaporation with Dr. Richard Kocan she has conducted studies examining sublethal pathological and genotoxic effects arising as a consequence of exposure of fish to environmental contaminants. Drs. Landolt and Kocan have also studied teratogenic effects that develop following exposure of fish embryos to pure compounds (e.g. benzo(a)pyrene) and complex mixtures (sea surface microlayer). For the past six years, she has collaborated with Dr.Jim Winton on studies of IHN virus and bacterial kidney disease. In addition to her professorial duties, Dr. Landolt has held several administrative posts. From 1983-1991 she was Associate Dean of the College of Ocean and Fishery Sciences. Since 1991 she has served as Director of the UW School of Fisheries.

Dr. Landolt is an experienced histopathologist whose expertise is frequently sought by regulatory agencies and other entities. She has participated in several large-scale field stucles examining the prevalence of idiopathic diseases in fish that reside in contaminated embayments. These studies have been sponsored by the US Environmental Protection Agency, the Municipality of Metropolitan Seattle and the Washington Department of Ecology. Currently, she is the histopathologist for the Puget Sound Ambient Monitoring Program, a study supported by the Washington Department of Fish and Wildlife . Because of her knowledge and experience, Dr. Landolt was asked by the Exxon Corporation and by Dames & Moore to evaluate flathead sole tissues and pink salmon alevins that were collected following the Exxon Valdes oil spill.

Dr. Landolt is a member of the American Fisheries Society Fish Health Section and is Associate Editor of the Journal of Aquatic Animal Health. She is an author on more than 50 scientific publications. Publications pertinent to the proposed study are listed below.

Key personnel (other than P.I.'s)

Dr. Tom Mehl. Res. Technologist II.

Ph.D. University of Washington Dept. of Pathology. Post-doctoral fellowships at University of Miami (Fla) and Univ. of Washington School of Medicine. Two years experience working with P.I. (Kocan) on EVOS projects in 1991- 1993. Experience as environmental and water quality chemist and laboratory rearing of various fish species. Extensive experience with statistical evaluation of experimental data.

Ms. Mary Bradley, Prof. Staff.

One year experience at Marrowstone Island maintaining saltwater facility with sterile seawater equipment. Experience with fish blood collection, fish diseases, necropsy, data collection and record keeping, use of Excel spread sheets, maintenance of u.v. sterilizers, carbon filters, sand filters fluid metering pumps and water chilling equipment. Previous experience with Washington State Dept. of Fisheries Shellfish Lab and Coast Oyster Co. (Quilcene, WA). Monitored seawater systems, heaters, filters, maintained algae cultures, quantitated shellfish larvae and cultured spawning oysters.

4/24/95 NUM

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Section III

Survival, Performance and Reproduction in the Pacific herring (Clupea pallasi)

Christopher J. Kennedy & Anthony P. Farrell

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PROJECT DESIGN

A. Objectives

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Given the above information base, it is not clear at this time whether VHSV, ITP, or oil exposure, or some combination causes a decline in herring survival, performance or reproductive fitness. These issues can be resolved with the three-tiered experimental approach outlined as components 1, 2 and 3 in the request for proposals:

- 1) Further field sampling; this corresponds to component #1 of the Request for Detailed Project Proposals.
- Testing of Koch's postulates for the roles of Viral Hemorrhagic Septicemia Virus and *lchthyophonus* in causing disease in PWS herring; this corresponds to component #2 of the Request for Detailed Project Proposals.
- 3) Laboratory experiments to document cause-effect and interactive relationships for oil, VHSV and ITP on herring survival, performance and reproduction and biological and abiotic factors such as density and temperature which may modify effects; this corresponds to components #2 and #3 of the Request for Detailed Project Proposals.

To execute this 3-tiered approach, a collaboration and integration of studies is necessary with Dr. Richard Kocan at the University of Washington and Dr. Gary Marty at University of Davis. It is also agreed that for the entire project to reach a successful endpoint it must span several years. For fiscal year 1995 it is further agreed that Dr. Marty performs field sampling and pathology, whereas Dr. Kocan works on setting up a disease-free herring stock and estimates the infection rate of local herring populations. Drs. Kennedy and Farrell's research focus will be studies on the acute and chronic effects of oil, VHSV and ITP exposure. We will use the same herring stocks as Dr. Kocan to ensure full integration of the studies in components 2 and 3 outlined in the request for proposals.

The endpoints that will be used to determine cause-effect relationships will be ecologically relevant stress responses as well as lethality. Conventional methods of evaluating stress to acuatic organisms often only examine one stress variable or a single level of organization and have been criticized as 'lacking ecological realism' (Cairns, 1981; NRC, 1981; Adams, 1990). The extrapolation of laboratory bioassays to the natural environment is difficult. It is therefore imperative to use ecologically relevant endpoints in laboratory-based bioassays. The review by Adams (1990) suggests a bioindicator approach to assessing stress as involving measurements of a suite of selected stress responses at several levels of biological organization ranging from the subcellular and biochemical levels to those at the ecosystem level. We will use such an approach to elucidate the causal relationship between potential stressors (oil contamination, VHSV and ITP) and their effects on herring. In the long-term we will examine four major ecologically relevant classes; 1) immunological fitness, 2) reproductive fitness, 3) physiological fitness and, 4) biochemical fitness.

The overall hypothesis being tested in this section of the proposal is: 'The exposure of herring to VHSV, ITP or oil or combinations of these parameters reduces herring fitness in one or more of the following categories: 1) immunology, 2) reproduction, 3) physiology, and 4) biochemistry.'

B. Methods

<u>Fish.</u> Disease-free young of the year from PWS will be raised by Dr. Kocan's group and should be available in approximately December 1995. At 5-6 g these juveniles will be suitable for sublethal toxicological testing and disease challenges. Our work (Johansen et al. 1995; Kennedy et al. in press) has successfully used rainbow trout of this size range in determining the sublethal toxicity of several natural wood products and antisapstain chemicals. As a fall back position, should the raising of herring from eggs fail, local (Washington) juvenile herring for which the background disease state will be determined will be used. Local adult herring for which the background disease state will be determined will be used to examine the effects on reproduction. Herring will be kept at the sea water facilities at Marrowstone Biological Station, Port Townsend, WA.

Exposure matrix. The experimental matrix (Figure 1) has seven (7) exposure cells and a control cell. The 3X3 design takes into account the three variables, oil contamination, VHSV and ITP singly or in various combinations. The exposures are: 1) VHSV only. 2) VHSV and ITP, 3) VHSV and oil, 4) ITP only, 5) ITP and oil, 6) oil only, 7) oil, VHSV, and ITP and 8) control fish which are pathogen-free and not exposed to any of the three variables. Each exposure cell will utilize approximately 40 fish. Statistics to be used will be performed by the statistics department at Simon Fraser University. This exposure scenario will allow the determination of the relevant parameter or combination which reduces herring fitness. These experiments will be performed in conjunction with the experiments performed by Dr. Kocan. VHSV and ITP exposures will be done simultaneously using predetermined doses (Dr. Kocan's group will determine these in component #2) of the pathogens. Dr. Kocan's group will examine disease parameters in these fish and our group will examine herring fitness as outlined previously. Fish will be exposed to oil using the dosing apparatus described in Johansen and Geen (1990) and fish examined for fitness or disease incidence by Dr. Kocan's group. We will begin our study with cells 1, 4 and 6 of Figure 1, which examine the effects of oil only, VHSV only and ITP only. When the effects of oil, VHSV and ITP on herring fitness are determined, the effects of the stressors of density and temperature will be determined. In all likelihood, some of the cells may be eliminated for the density and temperature studies. In these experiments, herring will be exposed to oil, VHSV and ITP under different densities and temperature regimes. Dr. Kocan's group will examine disease conditions in these experiments.

Figure 1. Various exposure scenarios and parameters. Superimposed upon this matrix are various doses and modulators such as density and temperature factors.

	VHSV	ITP	OIL	
VHSV	1. VHSV only	2. VHSV + ITP	3. VHSV + O	
ITP		4. ITP only	5. ITP + OIL	
OIL —			6. OIL only	

VHSV+ITP+OIL Controls

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<u>Fitness measurements.</u> Figure 2 illustrates the generic set of fitness tests and measurements that will be performed on Pacific herring following exposure to a given stress parameter. Four replicate trials will be performed for each exposure cell to test for each of the endpoints of 1) immunological fitness, 2) physiological fitness (by swimming performance), 3) biochemical fitness, and 4) reproductive fitness. In FY 96, all four parameters will be examined.

Figure 2. Generic fitness tests to be examined in each of the exposure scenarios mentioned in Figure 1.



In detail, the areas of fitness to be examined are:

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i) Immunological Fitness. Fish combat pathogenic microorganisms by an immune system which is comparable to other vertebrates. There is little direct evidence to link the contamination event of the Exxon Valdez with the increased occurrence of VHSV or ITP in herring in PWS. It has been shown that exposure to contaminants can affect the immune system of fish and compromise their ability to resist disease (Adams, 1990). It is also known that stress in general reduces disease resistance.

We will assess immunocompetence in fish following exposure to the stressors by measuring several immunological indicators, such as differential white blood cell counts, phagocyte activity using the nitroblue tetrazolium assay and glass adherent phagocytes, lysozyme assay using the lysoplate method, and antibody titers (IgG). Since it has been suggested by Meyers et al. (1993) that the progressive ulcerating skin lesions which occur in herring during an VHSV epizootic may act as portals of entry for secondary microbial infections, immunocompetence will also be measured by a disease challenge with the marine bacterium *Vibrio anguillarum* to determine the potential for a secondary infection. Methods for these measurements are described in Johansen et al. (1994) and Stolen et al. (1992).

ii) Physiological Fitness. Many stress-induced physiological events alter the capacity of fish to perform various physiological functions. Performance tests can be viewed as a form of bioassay that measures the capacity of fish to carry out essential life processes such as the ability to swim. These tests are particularly powerful tools for assessing stress as they incorporate several levels of biological organization and are therefore integrative in nature (Schreck 1990). In this section, we will examine the effects of the stressors on the swimming performance of herring. Ultimately swimming performance affects the ability of herring to forage and avoid predation.

One of the signs of VHSV infection in fish is lethargy and listlessness and frenzied swimming in circles at the terminal stages of disease. It is obvious that a reduced swimming performance may result reduced survival due to predation and an inability to secure food. Swimming involves the integrated effects of numerous physiological processes. Estimating maximum aerobic swimming ability can provide a sensitive index to general health and stress in fish and an index of the ability to avoid predation (Adams et al., 1990), since many physiological systems have to work maximally in a coordinated fashion.

Maximum aerobic swimming performance will be examined by determining the critical swimming speed of fish following exposure to oil, VHSV or ITP. In addition, schooling behavior will be noted. The assessment of swimming performance seems particularly relevant for the present study. ITP infection is high in both skeletal muscle and cardiac muscle of herring sampled from PWS (Freiberg and Farver, 1995), both of which are critical to swimming. It is likely that the ITP infection causes significant muscle tissue damage since high serum CPK levels correlate with ITP infection (Freiberg and Farver, 1995). We predict that cardiac ITP infection and damage will be particularly damaging to swimming performance and survival. Methods of determining swimming performance are described in Nikl and Farrell et al. (1991).

iii) Biochemical Fitness. A wide variety of molecular and biochemical responses to adverse environmental stimuli have been described for teleosts (Thomas 1990).
Biochemical alterations can be used as sensitive indicators of stress and show a more rapid response to environmental stressors than most other biological measurements. As well, measurements of molecular and biochemical indicators can often provide specific information on the nature of the stressor and its mechanism of action.
Biochemical parameters which have been shown to be good indicators of stress induced by contaminant exposure include: plasma cortisol, plasma glucose and lactate, leucocrit and hematocrit. We will measure these hematological variables following exposure to oil, VHSV and ITP. Analytical methods are described in Johansen et al. (1994).

The data from Freiberg and Farver (1995) indicate that measurements of creatine phosphokinase (CPK) in various tissues is highly correlated with fish lesions. In fish, CPK levels are elevated in ITP-infected herring indicating cellular damage in infected tissue. It is possible to measure CPK isoforms to identify the specific tissues damaged (CPK1, CPK2, CPK3: brain. cardiac and skeletal). Since we predict that cardiac tissue damage may have a proximate linkage to nerring survival, we will measure also measure these isoforms electrophoretically.

A contribution to the field sampling, component #1: Support services will be supplied to the analysis of the field samples in each year. The measurement of differential white blood cells is an indicator of the immunological status of fish and will be measured in blood smears sampled by Dr. Marty in PWS and SS. Due the strong statistical relationship between CPK and lesions in herring, the various isoforms of this enzyme will be measured in 100 field samples collected in PWS.

iv) Reproductive Fitness. Any stressor, including disease and contamination, that interferes with the process of reproduction at the individual or population level is likely to

affect the survival of that species in a habitat. Reproductive development is a continuous process and may be subject to the effects of environmental perturbations at several stages of an organisms life cycle. Through this development there are several parameters which may be useful indicators of reproductive 'fitness' in fish. In the proposed experiments, mature herring will be exposed to oil, VHSV and ITP. The following parameters in herring will be examined for possible effects; 1) sperm motility, 2) egg characteristics such as egg number, size, volume buoyancy, 3) if fertilization in the laboratory is successful: hatching characteristics such as percentage hatching, altered weight and length and. 4) survival of larvae to fry stage. These characteristics have been measured in herring from PWS and will establish cause-effect relationships between oil, VHSV and ITP and reproductive alterations under controlled laboratory conditions.

C. Contracts and Other Agency Assistance

BioWest Environmental Ltd. Vancouver, B.C. \$26,000 CPK & WBC on herring blood:

D. Location

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The experiments described will be performed in conjunction with Dr. Kocan's group at the Marrowstone Biological Station, Port Townsend, WA. PWS herring eggs will be collected in Alaska by Dr. Kocan and raised at Marrowstone Biological Station. Any procedures with animals will be authorized by Simon Fraser University's Animal Care Committee.

SCHEDULE

A. Measurable Project Tasks for FY 96

FY96: Exposures of juvenile herring to oil, VHSV and ITP only and analysis of measurements in each category of 1) biochemistry, 2) swimming performance, 3) immunology and 4) reproduction indices in mature herring. Data analysis and relevant statistics will begin on collected data. Analyze differential white blood cell counts and creatine phosphokinase isozymes in field samples collected by Dr. Marty's group. Annual progress report.

Project supervision: Dr. C.J. Kennedy: logistics, report writing, exper. supervision Project supervision: Dr. A.P. Farrell; exper. logistics, report writing; data interpret. Technician: A. Wood: exposures and fitness measurements; analysis of field samples Technician:unknown: fitness measurements, particularly reproduction Graduate student: unknown: exposures and fitness measurements; data analysis

FY97: Exposures of juvenile herring to combinations of oil, VHSV and ITP and analysis of measurements in each category of 1) biochemistry, 2) swimming performance, 3) immunology and 4) reproduction indices in mature herring. Begin exposures of herring under different density conditions. Completion of data analysis for FY96 data. Begin data analysis on collected data for FY97. Analyze differential white blood cell counts and creatine phosphokinase isozymes in field samples collected by Dr. Marty's group. Annual progress report.

B. Project Milestones and Endpoints

Oct. - Dec. '95 March '96 May '96 March - Sept. '96 Oct. - Dec. '96 Jan. - March. '97 March - Sept. '97 Oct. - Dec. '97 Jan. - June '98 June - Sept. '98 Aug. - Dec. '98 Establish initial SPF herring for later studies Establish SPF model for studying VHSV & *I. hoferi* Fulfil Koch's Postulates for VHSV in model system Fulfil Koch's Postulates for *I. hoferi* in model system Describe effects of chemical stressors on herring Describe effects of physical stressors on herring Collect and analyze blood samples from experimental fish Evaluate immune response in chemically stressed fish Evaluate immune response in physically stressed fish Evaluate immune response in fish with multiple infections

C. Project Reports

Preparation of manuscripts for peer reviewed publications will begin as studies are completed and sufficient data is available. Pre-prints and reprints of these will be forwarded to the Trustee Council and Chief Scientist as they are received.

Dec.	'95	Progress report for FY 95
Dec.	'96	Progress report for FY 96
Dec.	'97	Progress report for FY 97
Dec.	'98	Final report for FY 95 through FY 98

COORDINATION AND INTEGRATION OF RESTORATION EFFORT

Alaska Dept. of Fish and Game will contribute to this project by supplying a working platform for artificial spawning of herring in PWS. assistance in capturing and spawning the fish and transportation of embryos between the collection site, Cordova and the airport. The National Biological Service (NBS) will contribute Dr. Winton's salary as well as space and equipment. Statistical consultation (project design / data analyses) will be obtained through the UW Center for Quantitative Science. Computer services (data entry, data analysis, word processing) will be provided by SOF and NBS. Histological processing of tissue samples will be done through the UW Dept. of Pathology and histopathological evaluation of tissues from experimental infections and challenges will be conducted at SOF. Cell culture, virology and molecular biology facilities will be provided by NBS. Filtered seawater facilities for contaminant exposure studies are available at the Marrowstone Island Field Station (NBS), as is sterile (VHSV-free) seawater for *in vivo* virus studies. Filtered seawater facilities are also available at Friday Harbor Laboratories (UW).

ENVIRONMENTAL COMPLIANCE

The National Oceanic and Atmospheric Administration (NOAA) is the lead federal agency for National Environmental Policy Act (NEPA) compliance for this project. This project has been granted categorical exclusion because it is essentially a laboratory study without environmental consequences. Fish infected with pathogens will be housed in an approved government facility designed and approved for pathogen studies and all effluents will be decontaminated. Samples will be collected by ADF&G personnel under authority of a scientific collector's permit issued by the ADF&G. Permits needed for work in the State of Washington are granted by Wasnington Dept. of Fish & Game to the Univ. of Washington (R.M. Kocan, P.I.). Collection of herring eggs and 0-age herring in Puget Sound will be done under contract to the Mr. Charles Eaton (R.V. Kittiwake) under a scientific collector's permit issued to R.M. Kocan by the Washington Dept. of Fisheries. Animal Care Committee approval of the study has been granted at the Univ. of Washington. Studies conducted by Simon Fraser University (SFU) will be coordinated with both the Field and Laboratory components of this project. Interactions will involve S.F.U. evaluation of blood chemistry from PWS fish and laboratory infected fish. Some studies will be conducted by SFU personnel at the Marrowstone Island facility because of its isolation and containment features. Data will be continually reviewed and synthesized by all three groups (U.C. Davis, U of W and SFU).

PERSONNEL

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Dr. Kennedy is an Assistant Professor in the Department of Biological Sciences at Simon Fraser University. Dr. Kennedy has over 15 years experience in aquatic toxicology with special emphasis on fish biochemistry and physiology. He has strong research experience in subcellular, organismal and ecosystem level studies in aquatic toxicology as well as in analytical chemistry. He has produced 20 primary research publications and several reports under contract. As well, Dr. Kennedy has written two chapters on xenobiotics in the new book series "Biochemistry and Molecular Biology of Fishes".

Dr. Farrell is a Professor in the Department of Biological Sciences at Simon Fraser University. Dr. Farrell has extensive experience in fish physiology, aquatic toxicology and coordinating ecosystem level projects. He has produced over 100 primary research publications and several toxicology reports under contract. In addition, he is one of the editors for the world renowned treatise 'Fish Physiology" and edited a 300+ page report entitled "Towards Environmental Risk Assessment and Management of the Fraser River Basin".

Drs. Farrell and Kennedy have collaborated on several projects which have direct relevance to the proposed project with a central theme being the assessment of contaminant-induced stress on survival characteristics of fish. These projects were funded by Canadian Federal and Provincial Environmental agencies and include 'Biological Indicators of Stress in Fishes', 'Towards Criteria Development for Didecyldimethyl Ammonium Chloride', 'The Effects of Contaminants on Fish Reproduction" and "The Effects of Contaminants on Immunocompetence in Fish.' Christopher J. Kennedy, Ph.D. Dept. of Biological Sciences Simon Fraser University Burnaby, B.C. Canada V5A 1S6 pn (604) 291-5640 FAX (604) 291-3496 e-mail ckennedy@sfu.ca

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3/25/95 Version

October 1, 1995 - September 30, 1996

	Authorized	Proposed						****
Budget Category:	FFY 1995	FFY 1996						
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General Administration		\$14.0	FFY 1997	FFY 1998	FFY 1999	FFY 2000	FFY 2001	FFY 2002
Project Total	\$0.0	\$204.1			1	f		1
Full-time Equivalents (FTE)		0.1						
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Contractual Costs:			Proposed		
Description			FFY 1996		
Contract with non-tr	rustee agency Field (Marty)		53.9		
Contract with non-tr	rustee agency Lab (Kocan)		64.5		
Contract with non-trustee agency Challenge(Kennedy)					
Vessel Charter - Sitk	ca (R/V Medea, seiner, shipping)		7.1		
Vessel Charter - Cor	dova (Sampling vessel, shipping)		8.9		
When a non-trustee organ	nization is used, the form 4A is required.	Contractual Total	\$159.6		
Commodities Costs:			Proposed		
Description			FFY 1996		
Sampling supplies			1.0		
Pathology Lab suppl	ies/ virology, bacteriology		6.5		
		Commodition Total	\$75		
			\$7.5		
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October 1, 1995 - September 30, 1996

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Subtotal	\$0.0	\$45.0	Estimated	Estimated	Estimated	Estimated	Estimated	Estimated
Indirect		\$8.9	FFY 1997	FFY 1998	FFY 1999	FFY 2000	FFY 2001	FFY 2002
Project Total	\$0.0	\$53.9						
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Full-time Equivalents (FTE)		0.0						
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Other Resources					<u> </u>		<u> </u>	
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Pers	Personnel Costs:			Months	Monthly		Proposed
	Name	Position Description		Budgeted	Costs	Overtime	FFY 1996
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		statistics		0.0	2,613		0.0
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Contractual Costs:	Proposed
Description	FFY 1996
Necropsy; 160 at \$19	3.0
Histopathology; 160 at \$179	28.7
plasma chemistries; 160 at \$17	2.7
Osmolalities 160 at \$3	0.5
IgM analysis 660 at \$6.67	4.4
Contractual Tota	\$39.3
Commodities Costs:	Proposed
Description	F FY 1996
Commodities Tota	\$0.0
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1						0.0
						0.0
						0.0
						0.0
						0.0
Thos	e purchases a	ssociated with re	eplacement equipment should be indicated by placement of an R.	New Ed	quipment Total	\$0.0
Exist	ing Equipment	Usage:			Number	
Desc	ription				of Units	
ł						
l						
1			•			
1						
Į						
					F	ORM 4B
	4000		Project Number: 96162 - interim		F	quinment
	1996		Project Title: Investigations of Herring Diseases in PWS - Field	d l		
			Name: IIC Davis - Marty			DETAIL
L		ł			L	I
		8 of 16				8/2/95

Budget Category:	Authorized	Proposed						
	FFY 1995	FFY 1996						
Personnel		\$44.9						
Travel		\$1.6						
Contractual		\$1.8						
Commodities		\$2.7						
Equipment		\$0.0		LONG	RANGE FUND	NG REQUIREM	ENTS	
Subtotal	\$0.0	\$51.0	Estimated	Estimated	Estimated	Estimated	Estimated	Estimated
Indirect		\$13.5	FFY 1997	FFY 1998	FFY 1999	FFY 2000	FFY 2001	FFY 2002
Project Total	\$0.0	\$64.5						
Full-time Equivalents (FTE)		8.0						
•			Dollar amoun	ts are shown in	thousands of	dollars.		
Other Resources								
1								

October 1, 1995 - September 30, 1996

Pers	onnel Costs:			Months	Monthly		Proposed
	Name	Position Description		Budgeted	Costs	Overtime	FFY 1996
							0.0
		personnel		4.0	9,037	C	36.1
		benefits		4.0	2,209		8.8
							0.0
							0.0
							0.0
							0.0
			고려가, 이상 가지 않는 것이 가지 않는 것을 했다. 이상 것이 아이지 않는 것 같아, 것을 받았다.				0.0
in Record The second							0.0
			영상에 가 성경했다.				0.0
							0.0
	L	1					0.0
		Subtota		8.0	11,246	(
			Tisland	Davie d	r Tatal	rersonnel lota	\$44.9
Irav			TICKEL Drice	Rouna	Total	Daily Daily	Proposed
	Description	on Contria / Anghorange total	Price	1 rips	Days	Per Dien	FFY 1996
	Heming review meeting - Koc	an - Seattle / Anchorage, total	1576	4	L L		1.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
						Travel Tota	\$1.6
		Project Number: 96162 - interim					FORM 4B
	1006	Project Title: Investigations of Herri	ng Diseases i	n PWS - Lab	Studies		Personnel
	1996 Project little: investigations of Herring Diseases in PWS - Lab Studies						
-	1000	Name: U of W - Kocan					& Travel

8/2/95

Contractual Costs:			Proposed
Description			FFY 1996
Graduate Operating for	ees		1.5
Services			0.3
		Contractual Total	\$1.8
Commodities Costs:			Proposed
Description			FFY 1996
Supplies			27
			2.7
		Commodities Total	\$2.7
	Project Numbers 06162 interim	F	ORM 4B
1996	Project Number: 90102 - Interim	Cor	ntractual &
1330	Nome: LL of M. Keeper	Co	mmodities
			DETAIL
· · · · · · · · · · · · · · · · · · ·	11 of 16		8/2/95

New Equipmen	Purchases:	Number	Unit	Proposed
Description		of Units	Price	FFY 1996
				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 purchas	s associated with replacement equipment should be indicated by placement of an R.	New E	quipment Total	\$0.0
Existing Equipr	ent Usage:		Number	
Description			of Units	
	Project Number: 96162 interim		l f	ORM 4B
1006	Project Number. 30102 - Interim		F	auipment
1330	Project little: Investigations of Herring Diseases - Lab Studies		,-	DETAIL
	Name: U of W Kocan			
			L	

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

	Authorized	Proposed						
Budget Category:	FFY 1995	FFY 1996						
Personnel		\$12.2						
Travel		\$2.5						이 가슴을 많다. 이 가슴을 많은 것을 들었다.
Contractual		\$1.1						
Commodities		\$4.2						
Equipment		\$1.5		LONG	RANGE FUND	ING REQUIREM	ENTS	
Subtotal	\$0.0	\$21.5	Estimated	Estimated	Estimated	Estimated	Estimated	Estimated
Indirect		\$3.7	FFY 1997	FFY 1998	FFY 1999	FFY 2000	FFY 2001	FFY 2002
Project Total	\$0.0	\$25.2						
·								
Full-time Equivalents (FTE)								
			Dollar amoun	ts are shown in	n thousands of	dollars.		a ana ana ang ang ang ang ang ang ang an
Other Resources						T	I	T
1996	Project Num Project Title Name: Sim	nber: 96162 : Investigation on Fraser Ur	! (formerly 9) ons of Herrin niv Kenne	6320 S) - In Ig Diseases ii Idy	terim Reques n PWS - Cha	st llenge		FORM 4A Non-Trustee DETAIL 8/2/95

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

Pers	onnel Costs:			Months	Monthly		Proposed
	Name	Position Description		Budgeted	Costs	Overtim	e FFY 1996
							0.0
	Wood	technician		4.0	1,833		7.3
		benefits		4.0	150		0.6
	Fisher	grad student		4.0	1,000		4.0
		benefits		4.0	80		0.3
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
				10.0			0.0
		Subtotal		16.0	3,063	Concernent Tests	
<u> </u>			T :	0	r Tatal	ersonner Tota	
Irav	el Costs:		LICKET	Kound	Total	Dail Des Dies	y Proposed
	Description		Price	inps	Days	Per Dien	I FFT 1996
	Vanaguuar ta Alagka (E tring	· 2 mostingel: Vencouver to Bort Townsone //	5 tring)				0.0
	Vancouver to Alaska (5 trips	, 2 meetings), vancouver to Fort Townsene (2.5
							0.0
		i					0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
					•	Travel Tota	I \$2.5
		P					
[FORM 4B
		Project Number: 96162 - interim				1	Personnel
	1996	Project Title: Investigations of Herrin	ng Diseases i	n PWS - Cha	llenge		& Travel
		Name: Simon Fraser Univ - Kennedy					

DETAIL

14 of 16

Contractual Costs:		Proposed
Description		FFY 1996
long distance, fax, postage, photocopy, etc		0.1
Blood sample analysis		1.0
	Contractual Total	\$1.1
Commodities Costs:		Proposed
Description		FFY 1996
Fish maintenaince; analytical reagents		4.2
·		
	Commodities Total	\$4.2
Project Number: 96162 interim	F	ORM 4B
1996 Project Title: Investigations of Herring Diseases in PM/S - Challenge	Co	ntractual &
Name: Simon Fraser Univ - Kennedy	Co	mmodities
15 of 16		DETAIL

New Equipment Purchases:	Number	Unit	Proposed
Description	of Units	Price	FFY 1996
			0.0
electric motors and output box 2 at \$750			1.5
			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 an R.	New E	uioment Total	\$1.5
Existing Equipment Usage:		Number	
Description		of Units	
1996 If of 16 Project Number: 96162 - Interim Project Title: Investigations of Herring Diseases in PWS - Cha	llenge	F	ORM 4B quipment DETAIL 8/2/95



APEX: Apex Predator Ecosystem Experiment in Prince William Sound and the Gulf of Alaska

Project Number: Restoration Category: Proposer: LeadTrustee Agency: Coperating Agencies: Duration: Cost FY 96: Cost FY 96: Cost FY 97: Cost FY 98: Cost FY 98: Cost FY 99: Cost FY 00: Cost FY 01 Cost FY 02 Geographic Area: Injured Resource/Service: 97163 96163 Research David Cameron Duffy, Project Leader. NOAA DOI, ADFG Five years \$1,982,600 \$1,964,000 \$1,964,000 \$1,500,000 \$ 700,000 \$000 \$000 Prince William Sound, Gulf of Alaska Common Murre, Pigeon Guillemot, Recreation and Tourism (candidate species: Black-legged Kittiwake)

ABSTRACT

Several seabird species such as Common Murre, Pigeon Gullemot, Marbled Murrelet and Black-legged Kittiwake have not recovered from the *Exxon Valdez* oil spill. This project tests the hypothesis that ecosystem-level changes in the food environment of Prince William Sound may be responsible for nonrecovery of seabirds. In addition, commercial fishing, subsistence, and tourism have been lost or reduced by the spill. Several other species such as Harbor Seal and Pacific Herring have not recovered. All of these may be affected by changes in the food environment. At a still larger scale, resoration efforts such as salmon hatcheries or enhanced fisheries may have effects at the ecosystem level that need to be identified, so that such efforts can be fine-tuned to ensure the long-term help and viability of Prince William Sound.

This study will use seabirds as probes of the trophic (foraging) environment of Prince William Sound and will compare their reproductive and foraging biologies, including diet, with similar measurements from the Barren Islands, an area with more suitable food. These measurements will be compared with hydroacoustic and net samples of fish to calibrate seabird performance with fish distribution and abundance. We will use fish samples to compare diet, energetics and reproductive parameters of the different forage-fish species to determine whether competitive and predatory interactions or different responses to the environment may be favoring the abundance of one fish species over another. Finally, we will use seabird performance, including diet, and stomach samples from predtaory fish such as cod and halibut to develop methodology for long-term monitoring of the recovery of Prince William Sound and for an overall understanding of how the Sound works, in conjunction with research efforts funded by the Trustee Council and other organizations.

INTRODUCTION

The spill from the oil tanker *Exxon Valdez* resulted in significant mortality of several seabirds and in acute massive damage to Prince William Sound (PWS) and the Gulf of Alaska (GOA). Five years

following the spill, several species have not recovered. This may be the result of lingering effects of the oil spill (toxicity of prey, sublethal effects of oil exposure to organisms, or enduring changes to ecosystem structure). On the other hand, other non-oil factors may be involved, such as predation, climate-driven ecosystem changes, or even 'random' perturbations.

Both to aid in the recovery of injured resources and to safeguard the long-term health of Prince William Sound, we need to understand the ecological processes that control the ecosystem. This project focuses on the trophic interactions of seabirds and the forage species they feed on. We chose food as the focus because: 1) much of seabird population theory and several empirical field tests have identified food as an important limiting factor; 2) seabird/fish researchers in the PWS/GOA complex have concluded that major changes in food have occurred during the period; 3) other factors such as oil toxicity and climate change might express themselves through the food supply; and 4) a knowledge of the forage food base is critical for other apex predators, such as marine mammals and predatory fish, as well as for any larger effort to manage the marine resources Prince William Sound in a sustainable manner.

We propose to study the distribution and abundance of prey species through acoustic sampling in relation to food, environmental conditions and possible competitors, then to examine the physical, behavioral and competitive limits to access to these forage species for seabirds. We will examine the reproductive consequences of such limitations for pigeon guillemots *Cepphus columba*, black-legged kittiwakes *Rissa tridactyla*, tufted puffins *Lunda cirrhata*, common murres *Uria aalge*, and predatory fish. By examining the diet and reproductive consequences for a surface-feeder (kittiwake), a benthic diver (pigeon guillemot), two pelagic divers (puffin and murre), and large fish, we should be able to build up a picture of the forage base for the entire seabird community, setting the stage for a long-term, low-cost monitoring program.

NEED FOR THE PROJECT

A. Statement of Problem

Numerous seabird species have declined between surveys in the 1970's and the 1990's in Prince William Sound: cormorant spp., kittiwake, glaucous-winged gull, Arctic tern, Kittlitz's and marbled murrelets, tufted and horned puffin, and pigeon guillemot. Colony trends for kittiwakes have been inconsistent with changes in total numbers, although kittiwake productivity has dropped between 1984 -1989 and 1990 - 1993. The population of pigeon guillemots in PWS has decreased from about 15,000 in the 1970's to about 3,000 in 1993. Based on censuses taken around the Naked Island complex, pre-spill counts were roughly twice as high as post-spill counts. Pigeon guillemots are listed as "Not recovering" in the 1994 Exxon Valdez Oil Spill Restoration Plan.

Common murres were among the species most damaged by the oil spill, but most of the oiled birds nested outside PWS. Murres are also listed as "Not recovering" in the 1994 Exxon Valdez Oil Spill Restoration Plan, although their reproduction appears normal.

The best evidence for a shift in trophic resources for seabirds within Prince William Sound comes from pigeon guillemots. No long-term diet data sets exist for other species or, like black-legged kittiwakes, diet exhibits great year to year variability. In 1994, sand lance accounted for only about 1% of prey items fed to guillemot chicks at Jackpot Island and about 8% at Naked Island; in contrast, in 1979 the sand lance component at Naked Island was about 55%. Gadids were much more prevalent in the diet of guillemot chicks on Naked Island in 1994 (ca. 30%) than they were in 1979-1981 (<7%).

Pre-spill studies of pigeon guillemots breeding at Naked Island suggest that sand lance are a preferred prey during chick-rearing. Breeding pairs that specialized on sand lance tended to initiate nesting attempts earlier and produce chicks that grew faster and fledged at higher weights than did breeding pairs that preyed mostly upon blennies and sculpins, at least in years when sand lance were readily available. Consequently, the overall productivity of the guillemot population was higher when sand lance were available.
The decline in the prevalence of sand lance in the diet of guillemots breeding at Naked Island might be a key element in the failure of this species to recover from the oil spill. The schooling behavior of sand lance, coupled with their high lipid content relative to that of gadids and nearshore bottom fish, might make this species a particularly high-quality forage resource for PWS pigeon guillemots. This is consistent with the observation that other seabird species (e.g., puffins, murres, kittiwakes) experience enhanced reproductive success when sand lance are available.

B. Rationale

Both scientific theory and common sense suggest that ecosystems change over time and that changes to one species or other component of the ecosystem may reverberate through the entire ecosystem. Climate variations, fishing, or an oil spill may trigger changes that can take years to become apparent. Similarly, restoration efforts following the *Exxon Valdez* oil spill might increase injured species that are predators or competitors of other injured species, preventing recovery. By studying only the species level, we may miss such effects. An ecosystem approach, such as the APEX study of the upper-trophic level predators of Prince William Sound, is designed to look for such indirect links and to give us an understanding of the ecological context lacking from much of the Council's earlier single-species work. In conjunction with Sound Ecology Project and the Nearshore Vertebrate Project, APEX will give us a basic understanding of the ecological processes that may affect future changes and restoration efforts in the Sound and will help us determine when we have finally restored a sustainable and healthy Prince William Sound.

C. Summary of Major Hypotheses and Objectives

Our objectives are to 1. understand the relation between forage fish and seabird populations; 2. to understand what factors determine the relative abundance of forage species, and 3. to develop a long-term monitoring effort for the Sound. These objectives are expressed through a series of hypotheses.

General hypothesis:

A shift in the Prince William Sound marine trophic structure has prevented recovery of injured resources.

Working Hypotheses

- 1. The trophic structure of PWS has changed at the decadal scale
- 2. Planktivory is the factor determining abundance of the preferred forage species of seabirds
- 3. Forage species differ in their spatial responses to oceanographic processes
- 4. Productivity and size of forage species change the energy potentially available for seabirds
- 5. Forage fish characteristics and interactions among seabirds limit availability of seabird prey
- 6. Seabird foraging group size and species composition reflect prey patch size
- 7. Seabird diet composition and amount reflect changes in the relative abundance and distribution of forage fish at relevant scales around colonies
- 8. Changes in seabird reproductive productivity reflect differences in forage fish abundance as measured in adult seabird foraging trips, chick meal-size and chick provisioning-rates
- 9. Seabird reproductive productivity is determined by differences in forage fish nutritional quality
- 10. Seabird species within a community react predictably to different prey bases

Further development of these hypotheses and their breakdown into working hypotheses and tasks is presented in the appendices.

D. Completion Dates

By the end of 1995, we will have tested the major assumption that food is affecting or limiting recovery of several seabird species, through a comparison of Prince William Sound and the Barren Islands. In

1996 and 1997, we will undertake a further test of this between the two areas to assess interannual variability. At the end of the three years, we will have a minimal sample size of three to test for such variability between the two sites. If there is no variability between years or sites, or if variability is extreme, we would discontinue the study. However, we plan to continue through FY 2000 to obtain a sample size of six, the minimal size at which statistical power becomes reasonable. We plan the same timeline for comparison of diet and reproductive measures for seabirds at different sites within Prince William Sound.

For forage fish, we expect that three years of sampling (FY 95-97) through hydroacoustics will allow us to determine if trends in distribution and relative abundance of forage fish can be detected within the Sound and if these can be linked to seabird foraging and reproductive parameters and to diets of predatory fish. We will then reduce sampling levels to those that appear to give the most information per unit effort. For example, we may find that sampling within five kilometers of a kittiwake colony provides as much information as sampling within 40 km, or that single inshore samples in June for guillemot food are as effective as repeated samples.

We plan to use three years of data on forage fish energetics, diet and reproduction (FY 1996 - 1998) to assess between-year variability and to explore whether these can be linked to trends in hydroacoustic data. Thereafter, we will reduce sampling to a few selected variables for testing schemes for long-term monitoring.

By FY 1999, we expect to have a bird and fish monitoring scheme that can be field tested during FY 1999 and FY 2000 through calibration with ongoing projects. We then expect the monitoring to continue as a separate project, with a budget of \$100 K - 200 K, as a means of providing early warning of developing problems in Prince William Sound.

COMMUNITY INVOLVEMENT

Community involvement in this project will take several forms. First, charter boat captains and crews from several communities will be involved in collecting the diet samples from predatory fish, Second, we plan to seek student interns from spill-area communities to participate in the island-based seabird projects. This will allow a direct interchange of information between researchers and the communities. Second, we plan to support a student to ask communities to share their knowledge of past changes in distribution and abundance of seabirds and forage fish in the Sound, as well as their suggestions on why such changes occurred. These may help us develop further hypotheses that can be tested during our field work.

We also plan a series of winter visits to communities to explain what we are doing and to ask for suggestions on how our work can be improved or expanded. This will also help us recruit interns, with the cooperation of the elders and the schools.

FY 96 BUDGET

Personnel	\$ 625.8
Travel	\$ 61.8
Contractual	\$ 1,038.6
Commodities	\$ 74.1
Equipment	\$ 36.0
Subtotal	\$ 1,836.3
Gen. Admin.	\$ 146.3
Total	\$ 1,982.6

96163 Titles and Numbers

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PROJECT COMPONENT	TITLE
96163 A	Abundance and Distribution of Forage Fish and their Influence on Recovery of Injured Species
96163 B	Foraging of Seabirds
96163 C	Fish Diet Overlap Using Fish Stomach Content Analysis
96163 D	Distribution of Forage Fish as Indicated by Puffin Diet Sampling
96163 E	Black-legged Kittiwakes as Indicators of Forage Fish Availability
96163 F	Factors Affecting Recovery of Pigeon Guillemot Populations
96163 G	Diet Composition, Reproductive Energetics, and Productivity of Seabirds
96163 H	Proximate Composition and Energetic Content of Selected Forage Fish Species in PWS
96163 I	APEX Planning and Project Leader
96163 J	Barren Islands Seabird Studies
96163 K	Using Predatory Fish to Sample Forage Fish
96163 L	Historical Review of Ecosystem Structure in the PWS/GOA Complex and Abundance and Distribution of Forage Fish in the Barren Islands

1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

	Authorized	Proposed	Р	ROPOSED FI	FY 1996 TRUS	STEE AGENC	IES TOTALS	
Budget Category:	FFY 1995	FFY 1996	ADEC	ADF&G	ADNR	USFS	DOI	NOAĂ
			\$0.0	\$95.9	\$0.0	\$0.0	\$1,064.7	\$822.0
Personnel	\$356.8	\$625.8						
Travel	\$46.0	\$61.8						
Contractual	\$748.5	\$1,038.6						1984) han di sanakan Ngan
Commodities	\$56.9	\$74.1				C		양왕() (1) (1) (1) (1) (1) (1) (1) (1) (1) (1) (1)
Equipment	\$50.2	\$36.0		LONG R/	ANGE FUNDIN	NG REQUIRE	MENTS	
Subtotal	\$1,258.4	\$1,836.3	Estimated	Estimated	Estimated	Estimated	Estimated	Estimated
General Administration	\$95.4	\$146.3	FFY 1997	FFY 1998	FFY 1999	FFY 2000	FFY 2001	FFY 2002
Project Total	\$1,353.8	\$1,982.6	\$1,964.0	\$1,964.0	\$1,964.0	\$700.0	\$0 .0	\$0.0
Full-time Equivalents (FTE)	4.9	14.9						
			Dollar amounts	s are shown in	thousands of	dollars.		
Other Resources	\$0.0	\$0.0	\$0 .0	\$0.0	\$0.0	\$0.0	\$0.0	\$0.0
	orage isit prey	r .			•			
	Duciest	luck and a						CODM 2A

1996

Prepared 4/28/95

Project Number: 96163 Project Title: APEX Lead Agency: FORM 2A PROJECT DETAIL

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Budget Category:	Aumonzea	Proposed				a san inan Salah na	a dharann na chuirtean a san th	
	FFY 1995	FFY 1996						
Personnel	\$42.8	\$20.0						
Travel	\$9.0	\$6.9						
Contractual	\$460.8	\$655.7						
Commodities	\$1.0	\$0.0						
Equipment	\$3.5	\$0.0		LONG RA	NGE FUNDIN	G REQUIREN	MENTS	
Subtotal	\$517.1	\$682.6	Estimated	Estimated	Estimated	Estimated	Estimated	Estimated
General Administration	\$28.1	\$28.6	FFY 1997	FFY 1998	FFY 1999	FFY 2000	FFY 2001	FFY 2002
Project Total	\$545.2	\$711.2	\$700.0	\$700.0	\$700.0	\$700.0		
Full-time Equivalents (FTE)	0.6	0.3				N		
			Dollar amounts	s are shown in	thousands of	dollars.		
Other Resources]				
Comments: This project was	first funded as	a component	of the Eorage I	Fish Ecosyste	m Study (0416	3) then as the	ADEX project	(051624)
					, ,			

1996 EXXON VALDEZ TRUSTI DUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

PM Name Position Description Step Budgeted Costs Overtime * B. Wright Program Manager GS12/5 4.0 5,000 0 * B. Wright Program Manager GS12/5 4.0 5,000 0 * B. Wright Program Manager GS12/5 4.0 5,000 0 * B. Wright Subtotal Subtotal Subtotal Subtotal Subtotal 0 * Subtotal Subtotal Subtotal Subtotal Subtotal 0 0 * Those costs associated with program management should be indicated by placement of an * Personnel Total Personnel Total Travel Costs: Ticket Round Total Daily PM Description Price Trips Days Per Diem * Juneau to Anchorage 444 8 15 225	FFY 1996 20.0 0.0 0.0 0.0 0.0 0.0 0.0 0.0 0.0
* B. Wright Program Manager GS12/5 4.0 5,000 0 * B. Wright Program Manager GS12/5 4.0 5,000 0 * Subtotal Subtotal * 4.0 5,000 0 Those costs associated with program management should be indicated by placement of an * Personnel Total Travel Costs: Ticket Round Total Daily PM Description Price Trips Days Per Diem * Juneau to Anchorage 444 8 15 225	20.0 0.0 0.0 0.0 0.0 0.0 0.0 0.0 0.0
Subtotal 4.0 5.000 0 Those costs associated with program management should be indicated by placement of an *. Personnel Total Travel Costs: Ticket Round Total PM Description Price Trips Days * Juneau to Anchorage 444 8 15 225	0.0 0.0 0.0 0.0 0.0 0.0 0.0 0.0
Subtotal 4.0 5,000 0 Those costs associated with program management should be indicated by placement of an *. Personnel Total Travel Costs: Ticket Round Total PM Description Price Trips Days * Juneau to Anchorage 444 8 15 225	0.0 0.0 0.0 0.0 0.0 0.0 0.0
Subtotal Subtotal 4.0 5.000 0 and the second	0.0 0.0 0.0 0.0 0.0 0.0
Subtotal Subtotal 4.0 5,000 0 is Those costs associated with program management should be indicated by placement of an * Personnel Total Travel Costs: Ticket Round Total Daily PM Description Price Trips Days Per Diem * Juneau to Anchorage 444 8 15 225	0.0 0.0 0.0 0.0 0.0
Subtotal 4.0 5,000 0 Those costs associated with program management should be indicated by placement of an * Personnel Total Travel Costs: Ticket Round Total Daily PM Description Price Trips Days Per Diem * Juneau to Anchorage 444 8 15 225	0.0 0.0 0.0 0.0
Subtotal 4.0 5,000 0 Those costs associated with program management should be indicated by placement of an * Personnel Total Travel Costs: Ticket Round Total Daily PM Description Price Trips Days Per Diem * Juneau to Anchorage 444 8 15 225	0.0 0.0 0.0
Subtotal 4.0 5,000 0 Those costs associated with program management should be indicated by placement of an * Personnel Total Travel Costs: Ticket Round Total Daily PM Description Price Trips Days Per Diem * Juneau to Anchorage 444 8 15 225	0.0 0.0
Subtotal 4.0 5.000 0 Those costs associated with program management should be indicated by placement of an * Personnel Total Travel Costs: Ticket Round Total Daily PM Description Price Trips Days Per Diem * Juneau to Anchorage 444 8 15 225	0.0
Subtotal 4.0 5,000 0 Those costs associated with program management should be indicated by placement of an * Personnel Total Travel Costs: Ticket Round Total Daily PM Description Price Trips Days Per Diem * Juneau to Anchorage 444 8 15 225	
Subtotal4.05,0000Those costs associated with program management should be indicated by placement of an *.Personnel TotalTravel Costs:TicketRoundTotalDailyPM DescriptionPriceTripsDaysPer Diem*Juneau to Anchorage444815225	0.0
Subtotal4.05,0000Those costs associated with program management should be indicated by placement of an *Personnel TotalTravel Costs:TicketRoundTotalDailyPM DescriptionPriceTripsDaysPer Diem*Juneau to Anchorage444815225	0.0
Subtotal4.05,0000Those costs associated with program management should be indicated by placement of an *.Personnel TotalTravel Costs:TicketRoundTotalDailyPM DescriptionPriceTripsDaysPer Diem*Juneau to Anchorage444815225	0.0
Those costs associated with program management should be indicated by placement of an " Personnel total Travel Costs: Ticket Round Total Daily PM Description Price Trips Days Per Diem * Juneau to Anchorage 444 8 15 225	* ***
Travel Costs:TickelRoundTotalDailyPM DescriptionPriceTripsDaysPer Diem*Juneau to Anchorage444815225	\$20.0
PM Description Price Trips Days Per Diem * Juneau to Anchorage 444 8 15 225	Proposed
* Juneau to Anchorage 444 8 15 225	FFY 1996
	6.9
	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 *	
These dosis associated with program management should be indicated by placement of an	\$6.9

1996	Project Number: 96163A Project Title: APEX/Forage Fish Assessment Agency: NOAA	FORM 3B Personnel & Travel DETAIL
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4/28/95

Contractual Co	sts:								Proposed
Description									FFY 1996
Forage Fish Ass	sessment Cont	tract							655.7
When a non-trus	slee organizati	ion is used, the	form 4A is re	equired.				Contractual Total	\$655.7 Proposed
Description									FFY 1996
				,					
L							(commodities Total	\$0.0
1996		Project Project Agency:	Number: Title: NOAA	96163A APEX/Fora	age Fish	Assessmen	t	F Coi Coi	ORM 3B ntractua 1 & nmoditie
	4 of 81	L							

New Equipment Purchases:	Number	Unit	Proposed
Description	of Units	Price	FFY 1996
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
Those purchases associated with replacement equipment should be indicated by placement of an R.	New Equ	ipment Total	\$0.0
Existing Equipment Usage:		Number	Inventory
			Agency
		i	
		ĺ	
		<u></u>	
Project Number: 96163A		F	ORM 3B
1996 Project Title: APEX/Forage Fish Assessment		Eq	uipment
Agency: NOAA			DETAIL

	Authorized	Proposed				1999 - Jacon Million Service († 19		· · · · · · · · · · · · · · · · · · ·
Budget Category:	FFY 1995	FFY 1996						
Personnel		\$123.0						
Travel		\$38.6						
Contractual		\$288.0						
Commodities		\$5.0						
Equipment		\$9.3		LONG R	ANGE FUNDI	NG REQUIRE	MENTS	
Subtotal		\$463.9	Estimated	Estimated	Estimated	Estimated	Estimated	Estimated
Indirect		\$191.8	FFY 1997	FFY 1998	FFY 1999	FFY 2000	FFY 2001	FFY 2002
Project Total	\$460.8	\$655.7	\$650.0	\$650.0	\$650.0			
				· · · · · · · · · · · · · · · · · · ·				
Full-time Equivalents (FTE)		21.0						
			Dollar amount	s are shown ir	thousands of	dollars.		
Other Resources				51				
			·					

Per	sonnel Costs:			Months	Monthly		Proposed
	Name	Position Description		Budgeted	Costs	Overtime	FFY 1996
	L. Haldorson	PI		4.0	8,000	0	32.0
	A. Paul	fish biologist		2.0	7,000	0	14.0
	K. Coyle	fish biologist		10.0	5,000		50.0
	J. McDonald	fish biologist		2.0	6,000		12.0
	P. Shoemaker	fish biologist		3.0	5,000		15.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
		Subiolal		21.0	31,000	0	*****
					Per	sonnel lotal	\$123.0
Tray	vel Costs:		Ticket	Round	Total	Daily	Proposed
	Description		Price	Trips	Days	Per Diem	FFY 1996
	Fairbanks to Juneau		570	4	8	265	4.4
	Seward to Juneau		/00	.8	8	265	1.1
	Juneau Io Anchorage		444	4	6	265	3.4
	Seward to Anchorage		700	4	8	265	4.9
	Fairbanks to Anchorage		410	4	8	265	3.8
	Seallie to Anchorage		1,112	4	12	200	/.0 C 9
	r di Daliks IV Seallie		1,200	3	12	205	0.0
							0.0
							0.0
							0.0
							0.0
1			LI			Travel Total	\$38.6
Ľ							

		And the second s
1996	Project Number: 96164A Project Title: APEX/Forage Fish Assessment Name: University of Alaska Fairbanks	FORM 4B Personnel & Travel DETAIL
		1

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Contractual Costs:			Proposed
Description			FFY 1996
vessel charter @ \$3,000 per da	ay x 65 days		195.0
Subcontract to Biosonics			93.0
		Contractual Total	\$288.0
Commodities Costs:		Connactual Folds	Proposed
Description			FFY 1996
Laboratory, field and office	e supplies		5.0
· · · · · · · · · · · · · · · · · · ·			
	•	Commodities Total	\$5.0
[]		F	ORM 4B
	Project Number: 96163A	Co	ntractua
1996	Project Title: APEX/Forage Fish Assessment		1 &
	Name: University of Alaska Fairbanks	Coi	mmoditie
			<u> </u>

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1996 EXXON VALDEZ TRUST. UNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

New Equipment Purchases:	Number	Unit	Proposed
Description	of Units	Price	FFY 1996
mid-water trawl	1	2,500	2.5
small purse seine	1	6,000	6.0
gigabyte hard drive	1	800	0.8
			0. 0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
Those purchases associated with replacement equipment should be indicated by placement of an R.	New Equ	ipment Total	\$9.3
Existing Equipment Usage:		Number	
Description		of Units	
1996 Name: University of Alaska Fairbanks	ıt	H Ec	FORM 4B quipment DETAIL

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	Authorized	Proposed						
Budget Category:	FFY 1995	FFY 1996						
Personnel	\$56.4	\$104.9						
Travel	\$6 .2	\$8.8						
Contractual	\$3.0	\$5.2						
Commodities	\$1.3	\$1.3			ganda (k. 1911) angan angan			
Equipment	\$7.7	\$2.4		LONG RA	NGE FUNDIN	G REQUIREN	AENTS	
Subtotal	\$74.6	\$122.6	Estimated	Estimated	Estimated	Estimated	Estimated	Estimated
General Administration	\$8.7	\$16.1	FFY 1997	FFY 1998	FFY 1999	FFY 2000	FFY 2001	FFY 2002
Project Total	\$83.3	\$138.7	\$139.0	\$139.0	\$139.0			
Full-time Equivalents (FTE)		2.3						and a second second
			Dollar amount:	s are shown in	thousands of	dollars.		
Other Resources								
			-				2700 common - di 270 provinsi s ^{idan}	

October 1, 1995 - September 30, 1996

Per	rsonnel Costs:		GS/Range/	Months	Monthly		Proposed
PM	Name	Position Description	Step	Budgeted	Costs	Overtime	FFY 1996
	B. Ostrand	PI	GS11/3	12.0	4,750		57.0
		Bio. Tech.	GS5	5.0	2,400		12.0
		Research Assistant (grad. student)		5.0	3,000		15.0
	D. Irons	*Project Manager	GS12	2.0	5,500		11.0
		*Expediter	Grade4/2	3.0	3,300		9.9
							0.0
							0.0
	1						0.0
							0.0
							0.0
							0.0
							0.0
		Subtotal		27.0	18,950	0	44 - 1 - 1
Tho	se costs associated with pro	gram management should be indicated by	placement of	an *	Per	sonnel Total	\$104.9
Tra	vel Costs:		Ticket	Round	Total	Daily	Proposed
PM	Description		Price	Trips	Days	Per Diem	FFY 1996
	Anchorage to Whittier		100	6	12	225	3.3
	Anchorage/Cordova to Valo	lez	200	 (6	12	225	3.9
	emergency travel and trave	el to scientific meetings					1.6
							0.0
H							0.0
							0.0
		:					0.0
H							0.0
]						0.0
	1						0.0
							0.0
	<u> </u>						0.0
Tho	se costs associated with pro	gram management should be indicated by	placement of	<u>an *.</u>		Travel Total	\$8.8
-							

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Project Number: 96163B Project Title: APEX/Seabird Interactions Agency: DOI FORM 3B Personnel & Travel DETA1L

1996

Contractual Costs:			Proposed
Description			FFY 1996
safety training			2.5
emergency repair equ	lipment		0.5
equipment on mainter	nance		0.1
telephone, film proces	ssing, postage and freight, publication page charges		2.1
When a non-trustee organ	ization is used, the form 4A is required.	Contractual Total	\$5.2
Commodities Costs:			Proposed
Description			FFY 1996
Scientific supplies (fil	m, waterproof notebooks, guidebooks, charts)		0.1
rain gear, rubber bool	is, and gloves for 3 people		0.6
duplication costs			0.4
office supplies (comp	uter disks, paper, pens)		0.2
		Commodition Total	64.0
		commodities fotar	<u>۹۱.3</u>
		F	ORM 3B
	Project Number: 96163B		atractual
1996	Project Title: ADEY/Seabird Interactions		
	Agency: DOI		moditio
		L	

New	Equipment Purchases:		Number	Unit	Proposed
Des	cription		of Units	Price	FFY 1996
	binoculars		2	350	1.4
	data entry system upgrade				1.0
					0.0
					0.0
					0.0
					0.0
					0.0
					0.0
					0.0
					0.0
					0.0
					0.0
Tho	se purchases associated with re	eplacement equipment should be indicated by placement of an R	New Equi	oment Total	\$2.4
Fris	ting Equipment Usage:			Number	Inventory
Des	cription			of Units	Agency
					<u>y</u>
L				<u> </u>	
	[[]	
	P1	roject Number: 96163B		F	ORM 3B
	1996 P	roject Title: APEX/Seabird Interactions		Eq	uipment
		aency: 001			DETAIL
		gener: Dor			
L					

			KODAMANTANI					
	Authorized	Proposed						
Budget Category:	FFY 1995	FFY 1996						
Dessenat	£10.0	622.2						
Traval	\$10.0							
Contractual	\$5.0							
Commodities	\$0.0	\$0.0						2 - E
Equipment	\$0.0	\$0.0		LONG RA	NGE EUNDIN	C REOLIRE	MENTS	
Subtotal	\$0.0	\$0.0	Ectimated	Ectimated	Estimated	Estimated	Estimated	Estimated
General Administration	\$18.5	\$1.7 \$4.8	ESUMALEU	ESUMALEU	ESUMALEU FEY 1999	ESUMALEU EEY 2000	Estimated FEV 2001	ESUMALEU EEV 2002
Droject Total	\$1.5	\$4.0	\$56.0	\$56.0	\$56.0	1112000	1112001	1112002
Fillect Total	\$21.U	\$30.5	\$50.0	430.0	450.0			
Full-time Equivalents (FTE)		0.8						
		0.0	Dollar amount	s are shown in	thousands of	dollars		
Other Resources				S are shown in			F	
				and for and such to			1	L
1996 Prepart 28/95	Project Project / Agency:	Number: 9 Fitle: AF NOAA	96163C PEX/Fish D	viet Over	lap			FORM 3A AGENCY PROJEC T DETAJL

Per	sonnel Costs:		GS/Range/	Months	Monthly		Proposed
PM	Name	Position Description	Step	Budgeted	Costs	Overtime	FFY 1996
	M. Sturdevant	PI	GS9/6	8.0	3,400		27.2
	B. Wright	* Program Manager	GS12/5	1.0	5,000		5.0
							0.0
l							0.0
							0.0
1							0.0
							0.0
							0.0
l							0.0
							0.0
							0.0
							0.0
		Subtotal		9.0	8,400	0	
Ino	se costs associated with pro	gram management should be indicated by	placement of	an ".	Per	sonnel Total	\$32.2
Tra	vel Costs:		Ticket	Round	Total	Daily	Proposed
PM	Description		Price	Trips	Days	Per Diem	FFY 1996
	Juneau to Cordova		350	10	36	225	11.6
	Juneau to Anchorage		444	- 2	6	225	2.2
1	Juneau to Fairbanks		500	1	3	225	1.2
							0.0
							0.0
							0.0
							0.0
							0.0
1							0.0
							0.0
l							0.0
Tho	se costs associated with pro	oram management should be indicated by	nlacement of	an *		Travel Total	\$15 D
<u>L</u>		generic should be indicated by					φ13.0 j

1996

Project Number: 96163C Project Title: APEX/Fish Diet Overlap Agency: NOAA FORM 3B Personnel & Travel DETAIL

Contractual Costs:	Proposed
Description	FFY 1996
When a non-trustee organization is used, the form 4A is required. Contractual Total	\$0.0
Commodities Costs:	Proposed
Description	FFY 1996
sample bottles, formalin, and microscope work supplies	4.5
Commodities Total	\$4.5
1996 Project Number: 96163C Project Title: APEX/Fish Diet Overlap Agency: NOAA	ORM 3B ntractua l & mmoditie

New Equipment Purchases:		Number	Unit	Proposed
Description		of Units	Price	FFY 1996
				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 wit	h replacement equipment should be indicated by placement of an R.	New Equ	ipment Total	\$0.0
Existing Equipment Usage:		1	Number	Inventory
Description			of Units	Agency
				¥
		1		
	· · · · · · · · · · · · · · · · · · ·			
]	[1
	Project Number: 96163C		F	ORM 3B
1006	Project Mulber. JOIDJC		Ec	uipment
1990	Aconcy: NOAA			DETAIL
	Agency: NOAA			
			L	

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ſ	Authorized	Proposed		an a		orana alian alianga sara		
Budget Category:	FFY 1995	FFY 1996						
Personnel	\$30.0	\$66.6						
Travel	\$0.0	\$0.0						
Contractual	\$0 .0	\$0.0						
Commodities	\$0 .0	\$0.0		n na na serie da ser Esta da serie	ng sa Siginala a Cabiya na <u>Siginala s</u>	den en e		
Equipment	\$0 .0	\$0.0		LONG RA	NGE FUNDIN	IG REQUIRE	MENTS	
Subtotal	\$30.0	\$66.6	Estimated	Estimated	Estimated	Estimated	Estimated	Estimated
General Administration	\$4.5	\$10.0	FFY 1997	FFY 1998	FFY 1999	FFY 2000	FFY 2001	FFY 2002
Project Total	\$34.5	\$76.6	\$77.0	\$77.0	\$77.0			
Full-time Equivalents (FTE)		1.6						T. 1981.
			Dollar amount	s are shown in	n thousands of	dollars.		
Other Resources								
			-					
1996	Project Project '	Number: 9 Title: AP ADF&G	96163C PEX/Fish D	iet Over:	lap			FORM 3A AGENCY

1996 EXXON VALDEZ TRUS1 OUNCIL PROJECT BUDGET

Per	sonnel Costs	5:		GS/Range/	Months	Monthly		Proposed
PM	Name		Position Description	Step	Budgeted	Costs	Overtime	FFY 1996
	1		fisheries technician III		6.0	3,400	0	20.4
			fisheries technician III		6.0	3,400	0	20.4
1			fisheries technician III		6.0	3,400	0	20.4
1	PCN 116110	i	*Lib. I		0.3	5,530	0	1.7
I	PCN115103		* Program Manager, FBIV		0.5	7,432	0	3.7
1								0.0
								0.0
ł								0.0
1								0.0
1								0.0
1								0.0
								0.0
			Subtota		18.8	23,162	0	
Tho	se costs asso	ciated with pro	gram management should be indicated b	y placement of	an *.	Per	sonnel Total	\$66.6
Tra	vel Costs:			Ticket	Round	Total	Daily	Proposed
PM	Description	A 4 7		Price	Trips	Days	Per Diem	FFY 1996
l								0.0
								0.0
								0.0
								0.0
								0.0
1	1							0.0
l	1							0.0
								0.0
1								0.0
								0.0
							-	0.0
The	se costs asso	ciated with pro	oram management should be indicated b	v placement of	an *.		Travel Total	\$0.0
L				/				
		1						
			Project Number: 96163C					FORM 3B
	1996		Project Title: APFY/Fich	Diet Ove	rlan		P	ersonnel
			Adency: ADELC	DIEL UVE	riah		8	Travel
1			Ingency. Abras					DETAIL

1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

Contractual Costs:			Proposed
Description			FFY 1996
When a non-trustee organiza	ation is used, the form 4A is required.	Contractual Total	\$0.0
Commodities Costs:			Proposed
Description			FFY 1996
		••	
		Commodities Total	\$0.0
1996	Project Number: 96163C Project Title: APEX/Fish Diet Overlap Agency: ADF&G	F Coi Cor	ORM 3B htractua l & nmoditie s

1996 EXXON VALDEZ TRUSIEC COUNCIL PROJECT BUDGET

New Equipment Purchases:	Number	Unit	Proposed
Description	of Units	Price	FFY 1996
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
Those purchases associated with replacement equipment should be indicated by placement of an R.	New Equi	pment Total	\$0.0
Existing Equipment Usage:		Number	Inventory
Description		of Units	Agency
		1	
L]	
]	[]
Project Number: 96163C		F	ORM 3B
1996 Project Title: APEX/Fish Diet Overlap		Eq	uipment
Agency: ADF&G		1	DETAIL

	Authorized	Proposed			and a second		and good and the second second	
Budget Category:	FFY 1995	FFY 1996						
Personnel	\$10.0	\$31.0						
Travel	\$3.3	\$6.2						
Contractual	\$0.0	\$0.0						
Commodities	\$7.4	\$9.5						
Equipment	\$19.3	\$20.9		LONG RA	NGE FUNDIN	IG REQUIRE	MENTS	~
Subtotal	\$40.0	\$67.6	Estimated	Estimated	Estimated	Estimated	Estimated	Estimated
General Administration	\$1.5	\$4.7	FFY 1997	FFY 1998	FFY 1999	FFY 2000	FFY 2001	FFY 2002
Project Total	\$41.5	\$72.3	\$72.0	\$72.0	\$72.0			
	· · · · · · · · · · · · · · · · · · ·							
Full-time Equivalents (FTE)		0.9						
• • •		L	Dollar amounts	s are shown ir	thousands of	dollars.		
Other Resources								
Comments:								
					• .			
[<u>]</u>	1							FORM 3A
	Project	Number: 9	5163D					ACENCY
1996	Project	Title: AP	PEX/Puffin	is as Sami	plers			
	Agency:	DOI	•		•			PROJECT
								DETAIL
Prepared: 4/28/96							L	

Per	sonnel Costs:		GS/Range/	Months	Monthly		Proposed
PM	Name	Position Description	Step	Budgeted	Costs	Overtime	FFY 1996
	J. Piatt	PI	GS12/4	3.0	5,000		15.0
		project assistant	GS9	8.0	2,000		16.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
		Cubledel		11.0	7.000		0.0
The	a costs associated with pro-	Subioial	placement of		7,000		\$21.0
	se costs associated with pro	grant management should be indicated by	Tieket	an . Dound	Fei		
		······	Price	Round	Totar	Dally Bor Diam	Proposed
F M	Anchorage to Whittier		100	nips e	Uays 12	<u>225</u>	FFT 1990
	volunteer air faros		100	2	12	225	3.3
	float plane trips to study are		250		200	2	1.0
			230	۲	200	J	
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
Tho	se costs associated with pro-	gram management should be indicated by	placement of	an *.		Travel Total	\$6.2
							FORM 3B
	4000	Project Number: 96163D				D	arsonnel
	1996	Project Title: APEX/Puffin	ns as Samj	plers			Travol
		Agency: DOI				0	
							DETATE

Contractual Costs:			Proposed
Description			FFY 1996
When a non-trustee organizatio	n is used, the form 4A is required.	Contractual Total	\$0.0
Commodities Costs:			Proposed
Description			FFY 1996
food			3.5
tuel for skills			2.2
sleeping bags and pads	· · · ·		0.8
waterproof boat bags			0.2
rain gear and rubber boots			0.8
misc. camp gear (stoves, la	anterns, tools, cook kits)		1.0
scientific supplies (screens	, sampling bags, preservatives, scales)		1.0
		Commodities Total	\$9.5
1996	Project Number: 96163D Project Title: APEX/Puffins as Samplers Agency: DOI	F Co Co	FORM 3B ntractua 1 & mmoditie S

Nev	Equipment Purchases:		Number	Unit	Proposed
Des	cription		of Units	Price	FFY 1996
	SSB radio, antenna, battery		2	1,100	2.2
	VHF radios		2	500	1.0
	generator		1	500	0.5
	bomb shelter tents		4	620	2.5
	inflatable boat (Zodiak Mark	I or equivalent)	1	3,000	3.0
	outboard motors (25 h.p. an	d 15 h.p.)	1	3,000	3.0
	exposure suits		3	300	0.9
[binoculars		4	160	0.6
	burrow probe video system		1	6,200	6.2
1	climbing equipment		1	1,000	1.0
					0.0
					0.0
			L		0.0
Tho	se purchases associated with	replacement equipment should be indicated by placement of an R.	New Equi	pment Total	\$20.9
Exis	sting Equipment Usage:			Number	Inventory
Des	cription			of Units	Agency
		•••			
<u> </u>			<u></u>		
					1
		Project Number: 96163D		F	ORM 3B
	1996	Project Title: APEX/Puffins as Samplers		Eq	uipment
		Agency: DOI		1	DETAIL
		ingeney. Dor			

	Authorized	Proposed						
Budget Category:	FFY 1995	FFY 1996						
Personnel	\$54.2	\$102.7						
Travel	\$5.7	\$102.7						
Contractual	\$12.0	\$21.6						
Commodities	\$14.6	\$23.7						
Equipment	\$10.2	\$8.2	Eastallikadownae, and we and in [LONG RA	NGE ELINDIN	G REQUIREN	AENTS	en e e aestrado
Subtotal	\$96.7	\$164.0	Estimated	Estimated	Estimated	Estimated	Estimated	Estimated
General Administration	\$90.7	\$104.5		ESUITALEO	ESUITALEU	ESUMATED	ESUINALEU	
Broject Total	\$9.0	\$10.5 \$191.9	\$182.0	\$182.0	\$182.0	1112000	TT1 2001	FFT 2002
	\$105.7	\$101.0	\$102.0	\$102.0	\$102.0			
Eull time Equivalents (ETE)		2.2						
	I	2.5			thousends of	dollara		
Other Becaurees				s are snown in	i thousands of	donars.		
Other Resources	_ <u></u>							

Pers	sonnel Costs:		GS/Range/	Months	Monthly		Proposed
PM	Name	Position Description	Step	Budgeted	Costs	Overtime	FFY 1996
		PI	GS11	12.0	4,750		57.0
		Bio. Tech.	GS5	5.0	2,480		12.4
		Bio. Tech.	GS5	5.0	2,480		12.4
	D. Irons	* Project Manager	GS12	2.0	5,500		11.0
		*Expediter	Grade4/2	3.0	3,300		9.9
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
-		Subtotal		27.0	18,510	0	*
Ino	se costs associated with pro	gram management should be indicated by	placement of	an ".	Per	sonnel lotal	\$102.7
Tra	vel Costs:		Ticket	Round	Total	Daily	Proposed
PM	Description		Price	Trips	Days	Per Diem	FFY 1996
	Anchorage to Whittier to tra	nsport boat	1200	2	360	3	3.5
	Anchorage to Whittier		100	. 12			1.2
	float plane trips to study site		250	8			2.0
<u>]</u>	emergency travel and trave	to scientific meeting					2.0
							0.0
							0.0
	1						0.0
							0.0
							0.0
l							
Į							0.0
Tho	se costs associated with pro	oram management should be indicated by	nlacement of	I		Travel Total	\$8.7
		gram management should be indicated by	placement 0	GII .		TRAVET TUIDI	<u> </u>

1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

1996	Project Number: 96163E Project Title: APEX/Kittiwakes Agency: DOI	FORM 3B Personne & Travel DETAIL	\$ 21 1
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Contractual Costs:			Proposed
Description			FFY 1996
delivery of fuel and supplies	S		8.1
safety training for 3 people			3.0
truck rental			2.0
maintenance, cleaning, and	1 repair		8.5
When a non-trustee organization	n is used, the form 4A is required.	Contractual Total	\$21.6
Commodities Costs:			Proposed
Description			FFY 1996
food for 3 people for 120 d	ays		5.0
boat fuel: 150 gal/day for 6	0 days		12.0
scientific and camp supplie	s, software updates, and office supplies		6.7
4			
		Commodities Total	\$23.7
			ORM 3B
1006	Project Number: 96163E	Col	ntractua
1990	Project Title: APEX/Kittiwakes		1 &
	Agency: DOI	Cor	nmoditie
LJ			s

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1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

New Equipment Purchases:		Number	Unit	Proposed
Description		of Units	Price	FFY 1996
radio tags				6.2
emergency replacement eq	nuipment second s			2.0
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
I nose purchases associated wit	In replacement equipment should be indicated by placement of an R.	New Equi	pment lotal	\$8.2
Existing Equipment Usage:			Number	Inventory
Description			of Units	Agency
1	Project Number: 96163E	I	F	ORM 3B
1996	Project Title: APEX/Kittiwakes		Eq	uipment
	Agency: DOI			DETAIL
1	Agency. DOI			
			Lauran	

1996 EXXON VALDEZ TRUS . OUNCIL PROJECT BUDGET

October 1, 1995 - Juplember 30, 1996

	Authorized	Proposed		n ng sana na ng sana n S		and an entropy of the second se		
Budget Category:	FFY 1995	FFY 1996						
Personnel	\$71.8	\$120.3						
Travel	\$6.4	\$9.4						
Contractual	\$13.0	\$23.1						
Commodities	\$17.3	\$23.3		n a na sa	and and a start of the second seco	den sense sense de distancia de		
Equipment	\$7.0	\$2.0		LONG RA	NGE FUNDIN	IG REQUIREN	MENTS	
Subtotal	\$115.5	\$178.1	Estimated	Estimated	Estimated	Estimated	Estimated	Estimated
General Administration	\$11.7	\$19.7	FFY 1997	FFY 1998	FFY 1999	FFY 2000	FFY 2001	FFY 2002
Project Total	\$127.2	\$197.8	\$198.0	\$198.0	\$198.0			
Full-time Equivalents (FTE)	1.8	2.8						8 4 5 million and the second
			Dollar amounts	s are shown in	thousands of	dollars.		
Other Resources								
1996	Project I Project 5	Number: 9 Fitle: A	06163F APEX/Guill	emots				FORM 3A

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1996 EXXON	VALDEZ	TRUSTE	E COUNCIL	PROJECT	BUDGET
	October 1	1, 1995 - 3	September 3	30, 19 96	

-	Personnel Costs:			Months	Monthly		Proposed
PM	Name	Position Description	Step	Budgeted	Costs	Overtime	FFY 1996
	L. Hayes	PI	GS11	12.0	4,750		57.0
		Bio. Tech.	GS7	6.0	2,933		17.6
		Bio. Tech.	GS5	5.0	2,480		12.4
		Blo. Tech.	GS5	5.0	2,480		12.4
	D. Irons	*Project Manager	GS12	2.0	5,500		11.0
		*Expediter	Grade4/2	3.0	3,300		9.9
							0.0
							0.0
							0.0
	•						0.0
							0.0
							0.0
Subtotal				33.0	21,443	0	
Those costs associated with program management should be indicated by			placement of	an ".	Per	sonnel Total	\$120.3
Travel Costs:		Ticket	Round	lotal	Daily	Proposed	
PM	Description		Price	Irips	Days	Per Diem	FFY 1996
	Anchorage to Whittier to the	ansport boat	1200	2	480	3	3.8
	Anchorage to Whittier for 4 people		100	16			1.6
	float plane trips to study site		250	- 4			1.0
	travel to scientific meeting						2.0
							1.0
							0.0
							0.0
]				0.0
							0.0
							0.0
							0.0
Tho	Those costs associated with program management should be indicated by placement of an *.						

Contractual Costs:			Proposed			
Description						
delivery of fuel	and supplies		6.0			
safety training f	or 3 people		4.0			
truck rental			4.0			
maintenance, c	leaning, and repairs		7.1			
emergency equ	ipment repair		2.0			
When a non-trustee	organization is used, the form 4A is required	Contractual Total	\$23.1			
Commodities Cost			Proposed			
Description	3 . 		FFY 1996			
food for 3 peop	e for 120 days		4.0			
boat fuel: 150g	day for 60 days		12.0			
scientific and c	amp supplies, software updates, and office supplies		7.3			
		Commodities Total	\$23.3			
		F	ORM 3B			
4000	Project Number: 96163F	Col	ntractua			
1996	Project Title: APEX/Guillemots		1 &			
	Agency: DOI	Сог	mmoditie			
l i			~			

New Equipment Purchases: Num			Unit	Proposed				
Description			Price	FFY 1996				
emergency replacement equipment	t			2.0				
				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 replac	rement equipment should be indicated by placement of an R	New Equi	oment Total	\$2.0				
Existing Equipment Usage	ement equipment should be indicated by placement of an it.	I I I	Number	lavantaai				
Existing Equipment Osage:			of Lipits	Acency				
Description			01 01113	Agency				
		1	r)				
Proje	Project Number: 96163F		r Re	UNI 3D				
1996 Proje	ect Title: APEX/Guillemots	1						
Ageno	cy: DOI	ļ		DELAIL				
			L					
	Authorized	Proposed						
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Budget Category:	FFY 1995	FFY 1996						
Personnel	\$0.0	\$5.5					가 있다. 1993년 - 1993년 - 1993년 1993년 1993년 - 1993년 - 1993년	
Travel	\$0.0	\$0.0						
Contractual	\$148.4	\$168.4						
Commodities	\$0.0	\$0.0					Sector	
Equipment	\$0.0	\$0.0		LONG RA	NGE FUNDIN	G REQUIREN	AENTS	
Subtotal	\$148.4	\$173.9	Estimated	Estimated	Estimated	Estimated	Estimated	Estimated
General Administration	\$10.4	\$12.6	FFY 1997	FFY 1998	FFY 1999	FFY 2000	FFY 2001	FFY 2002
Project Total	\$158.8	\$186.5	\$187.0	\$187.0	\$187.0			
		<u></u>		· · · · · · · · · · · · · · · · · · ·				
Full-time Equivalents (FTE)	0.0	0.1						li Lawraig I.
			Dollar amounts	s are shown in	thousands of	dollars.		, <u>,</u>
Other Resources			I					
					1			

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October 1, 1995 - September 30, 1996

Personnel Costs:		GS/Range/	Months	Monthly		Proposed	
PM	Name	Position Description	Step	Budgeted	Costs	Overtime	FFY 1996
•	D. Irons	Project Manager	GS12	1.0	5,500		5.5
l							0.0
1							0.0
1							0.0
l I							0.0
1							0.0
							0.0
							0.0
							0.0
							0 .0
							0.0
į	<u> </u>						0.0
 		Subtotal	10000	1.0	5,500	0	
Tho	se costs associated with pro-	gram management should be indicated by	placement of	an *.	Per	sonnel Total	\$5.5
							3
Tra	vel Costs:		Ticket	Round	Total	Daily	Proposed
Tra PM	vel Costs: Description		Ticket Price	Round Trips	Total Days	Daily Per Diem	Proposed FFY 1996
Tra PM	vel Costs: Description		Ticket Price	Round Trips	Total Days	Daily Per Diem	Proposed FFY 1996 0.0
Tra PM	vel Costs: Description		Ticket Price	Round Trips	Total Days	Daily. Per Diem	Proposed FFY 1996 0.0 0.0
Tra PM	vel Costs: Description		Ticket Price	Round Trips	Total Days	Daily Per Diem	Proposed FFY 1996 0.0 0.0 0.0
Tra PM	vel Costs: Description		Ticket Price	Round Trips	Total Days	Daily Per Diem	Proposed FFY 1996 0.0 0.0 0.0 0.0
Tra PM	vel Costs: Description		Ticket Price	Round Trips	Total Days	Daily Per Diem	Proposed FFY 1996 0.0 0.0 0.0 0.0 0.0
Tra PM	vel Costs: Description		Ticket Price	Round Trips	Total Days	Daily Per Diem	Proposed FFY 1996 0.0 0.0 0.0 0.0 0.0 0.0 0.0
Tra PM	vel Costs: Description		Ticket Price	Round Trips	Total Days	Daily Per Diem	Proposed FFY 1996 0.0 0.0 0.0 0.0 0.0 0.0 0.0 0.0
Tra PM	vel Costs: Description		Ticket Price	Round Trips	Total Days	Daily Per Diem	Proposed FFY 1996 0.0 0.0 0.0 0.0 0.0 0.0 0.0 0.0 0.0
	vel Costs: Description		Ticket Price	Round Trips	Total Days	Daily Per Diem	Proposed FFY 1996 0.0 0.0 0.0 0.0 0.0 0.0 0.0 0.0 0.0 0.
	vel Costs: Description		Ticket Price	Round Trips	Total Days	Daily Per Diem	Proposed FFY 1996 0.0 0.0 0.0 0.0 0.0 0.0 0.0 0.0 0.0 0.
	vel Costs: Description		Ticket Price	Round Trips	Total Days	Daily Per Diem	Proposed FFY 1996 0.0 0.0 0.0 0.0 0.0 0.0 0.0 0.0 0.0 0.
	vel Costs: Description	pram management should be indicated by	Ticket Price	Round Trips	Total Days	Daily Per Diem	Proposed FFY 1996 0.0 0.0 0.0 0.0 0.0 0.0 0.0 0.
Tra PM Tho	vel Costs: Description se costs associated with pro	gram management should be indicated by	Ticket Price Price	Round Trips an [•] .	Total Days	Daily Per Diem Travel Total	Proposed FFY 1996 0.0 0.0 0.0 0.0 0.0 0.0 0.0 0.0 0.0 0.

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October 1, 1995 - September 30, 1996

Contractual Costs:			Proposed
Description			FFY 1996
Co Contract with the University	y of Alaska Cooperative Research Unit.		168.4
When a non-trustee organizatio	n is used, the form 4A is required.	Contractual Total	\$168.4
Commodities Costs:			Proposed
Description			FFY 1996
	۰.		
		Commodities Total	\$0.0
1996	Project Number: 96163G Project Title: APEX/Seabird Energetics Agency: DOI	F Cor Cor	ORM 3B ntractua 1 & nmoditie S

New Equipment Purchases:		Number	Unit	Proposed
Description		of Units	Price	FFY 1996
				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 will	h replacement equipment should be indicated by placement of an R.	New Equi	pment Total	\$0.0
Existing Equipment Usage:			Number	Inventory
Description			of Units	Agency
	,			
			[
	Project Number: 96163G		F	ORM 3B
1996	Project Title: APEX/Seabird Energetics		Eq	uipment
	Agency: DOI		1	DETAIL
			L	J
		1		

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	Authorized	Proposed		way ya watar i	e nave o se é anea	n a se geogra de la defensa a N		
Budget Category:	FFY 1995	FFY 1996						
Personnel	\$0.0	\$65.7						
Travel	\$0.0	\$8.5						
Contractual	\$148.4	\$21.9						
Commodities	\$0.0	\$31.9	and the second sec	an An an Anna an Anna an Anna an	o no Nitroune - estilo no necesió	a an	and a state of the	an a
Equipment	\$0.0	\$25.1		LONG R	ANGE FUNDI	NG REQUIRE	MENTS	
Subtotal	\$148.4	\$153.1	Estimated	Estimated	Estimated	Estimated	Estimated	Estimated
Indirect (10%)	N.e	\$15.3	FFY 1997	FFY 1998	FFY 1999	FFY 2000	FFY 2001	FFY 2002
Project Total	\$148.4	\$168.4	\$168.0	\$168.0	\$168.0			
							l u C M de la La	
Full-time Equivalents (FTE)		0.0	L	a second and the second se				
			Dollar amount	s are shown ir	thousands of	dollars.		
Other Resources		· · · · · · · · · · · · · · · · · · ·			<u> </u>		L	
			-					
1996	Project Project Name: DO	Number: 9 Title: AF I	96162G PEX/Seabir	d Energe	tics			FORM 4A Non- Frustee

Pers	onnel Costs:			Months	Monthly		Proposed
	Name	Position Description		Budgeted	Costs	Overtime	FFY 1996
		graduate student					11.4
		graduate student					11.4
ţ		field crew leader					4.9
		field tech.				i	4.4
		field tech.					4.4
		field tech.					4.4
		benefits					10.4
		student tuition					14.4
							0.0
							0.0
		[0.0
							0.0
			Subtotal	0.0	0]	0	
					Per	sonnel Total	\$65.7
Trav	el Costs:		Ticket	Round	Total	Daily	Proposed
	Description		Price	Trips	Days	Per Diem	FFY 1996
	presentation at national me	etings	1,000	3			3.0
	Fairbanks to Anchorage		416	8		455	3.3
	perdiem				13	155	2.0
	train Portage to Whittier		18	8			0.1
	train Portage to Whittier for	2 venicles	/0	2			0.1
		······		I I		Traval Tatal	0.0
						Travel Total	<u>Φ</u> δ.5
		ſ		<u></u>		J	
							FORM 4B
	1000	Project Number: 90	5163G			Pe	ersonnel
	1330	Project Title: Al	PEX/Seabird Energ	etics			() () ()

Name: DOI

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& Travel DETAIL

Contractual Costs:			Proposed
Description			FFY 1996
project suppo			5.0
vessel charter			6.0
vehicle rental			3.9
maintenance			7.0
		Contractual Total	\$21.0
Commodities Costs:			Proposed
Description			FTUPUSEU FFY 1996
laboratory, boat, climbing a	nd camp supplies		20.9
mustang suits x 6			1.8
fuel for boat			9.2
l l			1
<u> </u>			
1			
		Commodities Total	\$31.9
	Project Numbers 061626		
1996	Project Number: 961636		itractua
1330	Project Title: APEX/Seabird Energetics		Ι&
	Name: DOI	Con	nmoditie
[]			

New Equipment Purchases:		Number	Unit	Proposed
Description		of Units	Price	FFY 1996
Boston Whaler, 17'		1	15,000	15.0
outboard motor, 40h.p.		1	4,500	4.5
weatherports		2	2,800	5.6
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
Those purchases associated with	n replacement equipment should be indicated by placement of an R.		pment i otal	\$25.1
Existing Equipment Usage:			Number	
Description			or Units	
· · ·				
r				
	Project Number: 061630		F	ORM 4B
1006	Project Nulliper: 901036		Ec	uipment
1550	Names DOI	1		DETAIL
	Name: DOI			
			B	

	Authorized	Proposed		n series and a series of the				
Budget Category:	FFY 1995	FFY 1996						
Personnel	\$0.0	\$5.0						
Travel	\$0.0	\$0.9						
Contractual	\$0.0	\$35.5						
Commodities	\$0.0	\$0.0						
Equipment	\$0.0	\$0.0		LONG RA	NGE FUNDIN	IG REQUIRE	MENTS	
Subtotal	\$0.0	\$41.4	Estimated	Estimated	Estimated	Estimated	Estimated	Estimated
General Administration	\$0.0	\$3.2	FFY 1997	FFY 1998	FFY 1999	FFY 2000	FFY 2001	FFY 2002
Project Total	\$0.0	\$44.6	\$45.0	\$45.0	\$45.0			
Full-time Equivalents (FTE)	0.0	0.1						
			Dollar amount	s are shown ir	n thousands of	dollars.		
Other Resources						I	[
Commonic: This project of the		nation will dat	ermine the ere	vimate compo	sition and end	rantic contest	of selected fo	rade fich
encoded in the EVOS study occur	a This is a prov	posal issued s	ubmitted unde	r the Broad Av		comont		age non
species in the EVOS study area		pusai issueu s	ubinitieu unde	I THE PLOAD A	gency Announ	cement.		
1								
			and an intervention of the second					
							r	-
	Project	Number: 9	96163H					FORM 3A
1006	Project '	Title: A	PEX/Proxi	mate Com	position	of		AGENCY
1330	Forage F	ish	-		-			PROJECT
	Agency:	NOAA						DETAIL
							L	
Prepared: 4/28/96	L			······································			l	

Per	sonnel Costs:		GS/Range/	Months	Monthly		Proposed
PM	Name	Position Description	Step	Budgeted	Costs	Overtime	FFY 1996
*	B. Wright	Program Manager	GS12/5	1.0	5,000		5.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
		Subtotal		1.0	5, 0 00	0	
Tho	se costs associated with pro	gram management should be indicated by	placement of	an *.	Per	sonnel Total	\$5.0
Tra	vel Costs:		Ticket	Round	Total	Daily	Proposed
PM	Description		Price	Trips	Days	Per Diem	FFY 1996
*	Juneau to Anchorage		444	1	2	225	0.9
							0.0
				· · · ·			0.0
ţ.							0.0
)]							0.0
							0.0
							0.0
							0.0
							0.0
A							0.0
i.							0.0
Į							0.0
Tho	se costs associated with pro	gram management should be indicated by	placement of	an *.		Travel Total	\$0.9
							FORM 3B
ł		Project Number: 96163H					argonnal
	1996	Project Title: Proximate (Compositio	on of For	age Fish	P	er sonner
1		Agency: NOAA	▲ · · · · · · · · · · · · · · · · · · ·		-	<u>ہ</u> ا	Travel
-						1	

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October 1, 1995 - September 30, 1996

Contractual Costs:			Proposed
Description			FFY 1996
proximate composition	contract		35.5
When a non-trustee organiz	zation is used, the form 4A is required.	Contractual Total	\$35.5
Commodities Costs:			Proposed
Description			FFY 1996
· · · ·	······································	Commodities Total	\$0.0
1996	Project Number: 96163H Project Title: APEX/Proximate Composition of Forage Fish Agency: NOAA	F Cor Cor	ORM 3B htractua 1 & nmodítie s

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October 1, 1995 - September 30, 1996

New Equipment Purchases:		Number	Unit	Proposed
Description		of Units	Price	FFY 1996
				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 wit	h replacement equipment should be indicated by placement of an R.	New Equ	ipment Total	\$0.0
Existing Equipment Usage:			Number	Inventory
Description			of Units	Agency
	ſ	1		
	Project Number: 96163H			
4000	Project Title: APEX/Proximate Composition	of		uinment
1220	Forage Fish			DETATI.
	Agency: NOAA			
			L	ł

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October 1, 1995 - September 30, 1996

	Authorized	Proposed						
Budget Category:	FFY 1995	FFY 1996						
Personnel	\$0.0	\$18.0						
Travel	\$0 .0	\$3.0						
Contractual	\$0.0	\$0.0						
Commodities	\$0.0	\$3.5				waxa a	alanda ay ang	
Equipment	\$0.0	\$0.0		LONG R	ANGE FUNDI	NG REQUIRE	MENTS	
Subtotal	\$0 .0	\$24.5	Estimated	Estimated	Estimated	Estimated	Estimated	Estimated
Indirect (45%)	\$0.0	\$11.0	FFY 1997	FFY 1998	FFY 1999	FFY 2000	FFY 2001	FFY 2002
Project Total	\$0.0	\$35.5	\$36.0	\$36.0	\$36.0			
					<u></u>	11. T. M.		
Full-time Equivalents (FTE)	0.0	3.6						
			Dollar amounts	s are shown in	thousands of	dollars.		
Other Resources								
			Υ.		· .			
[]				• • • • • • • • • • • • • • • • • • •			1 ,	

October 1, 1995 - September 30, 1996

Personnel Costs			Months	Monthly		Proposed
Name	Position Description		Budgeted	Cosis	Overtime	FFY 1996
G. Worthy	PI		3.6	5,000		18.0
				-		0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
	Subiotal		3.6	5,000	U	£10.0
				Per	sonnei Totai	\$18.0
Travel Costs:		licket	Rouna	Total	Dally	Proposed
				Days	Per Diem	FFY 1996
Texas to Anchorage		1,500	2			3.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
						0.0
					Travel Total	\$3.0
	[1	r	
	Project Number: 96163H			ľ		FORM 4B
1000	Project Title: APEX/Prox	imate Com	position	of	P	ersonnel
1990	Forage Fish				6	Travel
	Name: Mayag N(M Universit	F 1 F			1	DETAIL

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Contractual Costs:	Proposed
Description	FFY 1996
Contractual Total	\$0.0
Commodities Costs:	Proposed
Description	FFY 1996
laboratory and sample handling supplies	3.5
Commodities Total	\$3.5
1996 Project Number: 96163H Project Title: APEX/Proximate Composition of Forage Fish Name: Texas A&M University	FORM 4B ntractua l & mmoditie s

October 1, 1995 - September 30, 1996

New Equipment Burchasses	en die en fanne in die en de staar wat die staar in die staar werden die staar die staar die staar die staar we	Number	1 4	Dropocod
New Equipment Furchases.		of Unite	Unit	Proposed
			FIICE	FFT 1990
				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	h replacement equipment should be indicated by placement of an R.	New Equ	ipment Total	\$0.0
Existing Equipment Usage:			Number	
Description			of Units	
1996	Project Number: 96163H Project Title: APEX/Proximate Composition Forage Fish Name: Texas A&M University	of	F	ORM 4B uipment DETAIL

	Authorized	Proposed	E States and the states of the					
Budget Category	FEY 1995	FFY 1996						
Dudget dutegery.	1111000							
Personnel	\$23.7	\$10.5						옷에서 모양을 깨끗했다.
Travel	\$2.4	\$0.0						
Contractual	\$94.3	\$104.8						
Commodities	\$0.0	\$0.0						
Equipment	\$0.0	\$0.0	States and the second sec	LONG RA	NGE FUNDIN	IG REQUIRE	MENTS	
Subtotal	\$120.4	\$115.3	Estimated	Estimated	Estimated	Estimated	Estimated	Estimated
General Administration	\$10.2	\$8.9	FFY 1997	FFY 1998	FFY 1999	FFY 2000	FFY 2001	FFY 2002
Project Total	\$130.6	\$124.2	\$115.0	\$115.0	\$115.0			
Full-time Equivalents (FTE)		0.2						
			Dollar amounts	s are shown in	n thousands of	dollars.		
Other Resources			Ι					
Comments: This component	of the APEX or	niect will provi	de scientific ou	ersight coord	tination nerto	mance tracki	and integra	tion of
			·					
1006	Dreisch							

Per	sonnel Costs:		GS/Range/	Months	Monthly		Proposed
PM	Name	Position Description	Step	Budgeted	Costs	Overtime	FFY 1996
*	D. Irons	Project Manager	GS12/5	1.0	5,000		5.0
•		Program Manager	GS12	1.0	5,500		5.5
							0.0
ļ							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
<u> </u>		Subtotal		2.0	10,500	0	
Tho	se costs associated with pro	gram management should be indicated by	placement of	an ".	Per	sonnel Total	\$10.5
Tray	vel Costs:	······································	Ticket	Round	Total	Daily	Proposed
<u>PM</u>	Description		Price	Trips	Days	Per Diem	FFY 1996
							0.0
				· · ·			0.0
							0.0
]						0.0
							0.0
l							0.0
8							0.0
							0.0
]						0.0
ll –							0.0
I							0.0
Tho	se costs associated with pro	oram management should be indicated by	nlacement of	LI an *	<u></u>	Travel Total	\$0.0
	to coold associated with pro	grant management should be indicated by	pidoonion or				

1996	Project Number: 96163I Project Title: APEX/Project Management Agency: DOI	FORM 3B Personnel & Travel
	Agency: DOI	& Travel DETAIL

Contractual Costs:	Proposed
Description	FFY 1996
contract with University of Alaska (use memorandum of understanding to transfer funds)	104.8
When a non-trustee organization is used, the form 4A is required Contractual Total	\$104.8
Commodities Costs:	Proposed
Description	FFY 1996
Commodities Total	\$0.0
1996 Project Number: 96163I Project Title: APEX/Project Management Agency: DOI Co	FORM 3B ntractua 1 & mmoditie

New Equipment Purchases:		Number	Unit	Proposed
Description		of Units	Price	FFY 1996
				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 wit	h replacement equipment should be indicated by placement of an R.	New Equi	pment Total	\$0.0
Existing Equipment Usage:			Number	Inventory
Description			of Units	Agency
	· · · · ·			
	r			1
1000	Project Number: 96163I		r	
1996	Project Title: APEX/Project Management]	Eq	
	Agency: DOI	Į		JETALL
			L	

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October 1, 1995 - September 30, 1996

Budget Category: Personnel Travel Contractual Commodities Equipment Subtotal Indirect (36.2%) Project Total Full-time Equivalents (FTE) Other Resources Comments: This component of results. The program managem	FFY 1995 \$58.2 \$5.0 \$5.0 \$1.0 \$0.0 \$69.2 \$25.1 \$94.3	FFY 1996 \$66.0 \$7.0 \$5.0 \$1.0 \$0.0 \$79.0 \$25.8 \$104.8 1.0	Estimated FFY 1997 \$105.0	LONG R Estimated FFY 1998 \$105.0	ANGE FUNDI Estimated FFY 1999 \$105.0	NG REQUIRE Estimated FFY 2000	MENTS Estimated FFY 2001	Estimated FFY 2002
Personnel Travel Contractual Commodities Equipment Subtotal Indirect (36.2%) Project Total Full-time Equivalents (FTE) Other Resources Comments: This component of results. The program managem	\$58.2 \$5.0 \$5.0 \$1.0 \$0.0 \$69.2 \$25.1 \$94.3	\$66.0 \$7.0 \$5.0 \$1.0 \$0.0 \$79.0 \$25.8 \$104.8 1.0	Estimated FFY 1997 \$105.0	LONG R Estimated FFY 1998 \$105.0	ANGE FUNDI Estimated FFY 1999 \$105.0	NG REQUIRE Estimated FFY 2000	MENTS Estimated FFY 2001	Estimated FFY 2002
Personnel Travel Contractual Commodities Equipment Subtotal Indirect (36.2%) Project Total Full-time Equivalents (FTE) Other Resources Comments: This component of results. The program managem	\$58.2 \$5.0 \$5.0 \$1.0 \$0.0 \$69.2 \$25.1 \$94.3	\$66.0 \$7.0 \$5.0 \$1.0 \$0.0 \$79.0 \$25.8 \$104.8 1.0	Estimated FFY 1997 \$105.0	LONG R Estimated FFY 1998 \$105.0	ANGE FUNDI Estimated FFY 1999 \$105.0	NG REQUIRE Estimated FFY 2000	MENTS Estimated FFY 2001	Estimated FFY 2002
Travel Contractual Commodities Equipment Subtotal Indirect (36.2%) Project Total Full-time Equivalents (FTE) Other Resources Comments: This component of results. The program managem	\$5.0 \$5.0 \$1.0 \$0.0 \$69.2 \$25.1 \$94.3	\$7.0 \$5.0 \$1.0 \$0.0 \$79.0 \$25.8 \$104.8 	Estimated FFY 1997 \$105.0	LONG R Estimated FFY 1998 \$105.0	ANGE FUNDI Estimated FFY 1999 \$105.0	NG REQUIRE Estimated FFY 2000	MENTS Estimated FFY 2001	Estimated FFY 2002
Contractual Commodities Equipment Subtotal Indirect (36.2%) Project Total Full-time Equivalents (FTE) Other Resources Comments: This component of results. The program managem	\$5.0 \$1.0 \$0.0 \$69.2 \$25.1 \$94.3	\$5.0 \$1.0 \$0.0 \$79.0 \$25.8 \$104.8 1.0	Estimated FFY 1997 \$105.0	LONG R Estimated FFY 1998 \$105.0	ANGE FUNDI Estimated FFY 1999 \$105.0	NG REQUIRE Estimated FFY 2000	MENTS Estimated FFY 2001	Estimated FFY 2002
Commodities Equipment Subtotal Indirect (36.2%) Project Total Full-time Equivalents (FTE) Other Resources Comments: This component of results. The program managem	\$1.0 \$0.0 \$69.2 \$25.1 \$94.3	\$1.0 \$0.0 \$79.0 \$25.8 \$104.8 1.0	Estimated FFY 1997 \$105.0	LONG R Estimated FFY 1998 \$105.0	ANGE FUNDI Estimated FFY 1999 \$105.0	NG REQUIRE Estimated FFY 2000	MENTS Estimated FFY 2001	Estimated FFY 2002
Equipment Subtotal Indirect (36.2%) Project Total Full-time Equivalents (FTE) Other Resources Comments: This component of results. The program managem	\$0.0 \$69.2 \$25.1 \$94.3	\$0.0 \$79.0 \$25.8 \$104.8 1.0	Estimated FFY 1997 \$105.0	LONG R Estimated FFY 1998 \$105.0	ANGE FUNDI Estimated FFY 1999 \$105.0	NG REQUIRE Estimated FFY 2000	MENTS Estimated FFY 2001	Estimated FFY 2002
Subtotal Indirect (36.2%) Project Total Full-time Equivalents (FTE) Other Resources Comments: This component of results. The program managem	\$69.2 \$25.1 \$94.3	\$79.0 \$25.8 \$104.8 	Estimated FFY 1997 \$105.0	Estimated FFY 1998 \$105.0	Estimated FFY 1999 \$105.0	Estimated FFY 2000	Estimated FFY 2001	Estimated FFY 2002
Indirect (36.2%) Project Total Full-time Equivalents (FTE) Other Resources Comments: This component of results. The program managem	\$25.1 \$94.3	\$25.8 \$104.8 1.0	FFY 1997 \$105.0	FFY 1998 \$105.0	FFY 1999 \$105.0	FFY 2000	FFY 2001	FFY 2002
Project Total Full-time Equivalents (FTE) Other Resources Comments: This component of results. The program managem	\$94.3	\$104.8 1.0	\$105.0	\$105.0	\$105.0	\$405 Q		
Full-time Equivalents (FTE) Other Resources Comments: This component of results. The program managem		1.0		20-1-1-1-1-1-1-1-1-1-1-1-1-1-1-1-1-1-1-1		\$105.U		
Full-time Equivalents (FTE) Other Resources Comments: This component of results. The program managem		1.0						
Other Resources Comments: This component of results. The program managem			State and an and a state of the					
Other Resources Comments: This component of results. The program managem			Dollar amount:	s are shown in	thousands of	dollars.		
Comments: This component of results. The program managem								
			,		•.			
				and the second				FORM 4A

Per	sonnel Costs:			Months	Monthly		Proposed
	Name	Position Description		Budgeted	Costs	Overtime	FFY 1996
	D. Duffy	PI		6.0	8,000		48.0
8. S		assistant PI		6.0	3,000		18.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
		Subtotal		12.0	11,000	0	
					Per	sonnel Total	\$66.0
Tra	vel Costs:		Ticket	Round	Total	Daily	Proposed
	Description		Price	Trips	Days	Per Diem	FFY 1996
	Anchorage to PWS commu	nities to complete information transfer					5.0
	Anchorage to Juneau		444	2	5	225	2.0
		,		、			0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
Ļ						Travel Total	\$7.0
				· · · · · · · · · · · · · · · · · · ·]		
					·		FORM 4B
1	1000	Project Number: 96163I				P	ersonnel
1	1220	Project Title: APEX/Proj	ect Manag	ement		r	Travel
1							TTUVCI

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Contractual Costs:			Proposed
Description			FFY 1996
data compatibility contract			5.0
		Contractual Total	\$5.0
Commodities Costs:			Proposed
Description			FFY 1996
supplies and phone			1.0
		Commodities Total	\$1.0
1996	Project Number: 96163I Project Title: APEX/Project Management Name: University of Alaska Anchorage	F Cor Cor	ORM 4B ntractua 1 & nmoditie s

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New Equipment Purchases:	Number	Unit	Proposed
Description	of Units	Price	FFY 1996
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
Those purchases associated with replacement equipment should be indicated by placement of an R.	New Equ	ipment Total	\$0.0
Existing Equipment Usage:		Number	
Description		of Units	
	1		
			100 g.t
		[
Project Number: 96163I		F	ORM 4B
1996 Project Title: APEX/Project Management		Eq	uipment
Name: University of Alaska Anchorage			DETAIL
		L	

	Authorized	Proposed		energe - Levends Latisticas y o		er and strength a service of	e andre state av	
Budget Category:	FFY 1995	FFY 1996						
Personnel	\$19.4	\$69.8						
Travel	\$6.4	\$2.3						
Contractual	\$3.4	\$10.6						
Commodities	\$3.8	\$4.8		a and an and	an. Babaa ka shi ka waxaa		lan in the	
Equipment	\$0.0	\$0.0		LONG RA	NGE FUNDIN	G REQUIRE	MENTS	
Subtotal	\$33.0	\$87.5	Estimated	Estimated	Estimated	Estimated	Estimated	Estimated
General Administration	\$3.1	\$11.2	FFY 1997	FFY 1998	FFY 1999	FFY 2000	FFY 2001	FFY 2002
Project Total	\$36.1	\$98.7	\$99.0	\$99.0	\$99.0			
Full-time Equivalents (FTE)	0.9	1.8						
			Dollar amount	s are shown ir	n thousands of	dollars.		
Other Resources								
Comments: This component is	designed to co	llect data on c	common murre	s, kittiwakes, a	and puffins on	the Barren Isl	ands (which is	in the
EVOS area) that will be used in	a multi-specie	es analysis of a	seabird produc	tivity and energy	rgetics.			
	-	-	-	-	-			
			•		÷			
· · · · · · · · · · · · · · · · · · ·	.							
	Project I	Number: 9	0103J	_		_		TUKM JA
1996	Project '	Fitle: AP	'EX/Barren	Islands	Murres a	nd		AGENCY
1000	 Kittiwake	26				1		I
-		20						PROJECT
	Agency:	DOI						PROJECT DETAIL

October	1,	1995	 September 	30,	1996
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Pers	sonnel Costs:		GS/Range/	Months	Monthly		Proposed
PM	Name	Position Description	Step	Budgeted	Costs	Overtime	FFY 1996
	D. Roseneau	PI	GS11/3	8.0	4,200		33.6
		camp leader/bio. tech.	GS7/2	8.0	2,900		23.2
		bio. tech	GS6/1	5.0	2,600		13.0
l							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
ł							0.0
							0.0
							0.0
		Subtotal		21.0	9,700	0	<u></u>
Tho	se costs associated with pro-	gram management should be indicated by	placement of	an *.	Per	sonnel Total	\$69.8
Trav	vel Costs:		Ticket	Round	Total	Daily	Proposed
PM	Description		Price	Trips	Days	Per Diem	FFY 1996
	meetings in Anchorage				10	225	2.3
				4			0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
					1		0.0
							0.0
L	L		L	L [0.0
Tho	se costs associated with pro-	gram management should be indicated by	placement of	an ".		Travel Total	\$2.3

1996Project Number: 96163J
Project Title: APEX/Barren Islands Murres and
Kittiwakes
Agency: DOIFORM 3B
Personnel
& Travel
DETAIL

Description FFY 1996 2 vessel days at \$2.1K/day 42 4 resupply fixed wing charter lights @ \$.25/trip 10 1 emergency helicopter trip 114 1 SCA volunteer in Homer, 3 months 114 1 SCA volunteer in Homer, 3 months 114 1 sceaning and repair of radios and outboard motors 115 2 monodities costs: 215 2
2 vessel days at \$2.1k/day 4.2 4 resupply fixed wing charter fights @ \$.25/trip 1.0 1 emergency helicopter trip 1.4 1 SCA volunteer in Homer, 3 months 3.4 cleaning and repair of radios and outboard motors 0.6 When a non-trustee organization is used, the form 4A is required. Contractual Total \$10.6 Zommodities Costs: Proposed Description FFY 1996 ood for field camp 3.1 ield, climbing, and camping gear 1.7 1.7 \$4.8
4 resupply fixed wing charter fights @ \$.25/trip 1.0 1 emergency helicopter trip 1.4 1 SCA volunteer in Homer, 3 months 3.4 cleaning and repair of radios and outboard motors 0.6 Mhen a non-trustee organization is used, the form 4A is required. Contractual Total Sommodities Costs: Proposed Description FFY 1996 od for field camp 3.1 ield, climbing, and camping gear 1.7 Commodities Total \$4.8
1 emergency helicopter trip 1.4 1 SCA volunteer in Homer, 3 months 3.4 cleaning and repair of radios and outboard motors 0.6 Alternative organization is used, the form 4A is required. Contractual Total Sommodities Costs: Proposed Description FFY 1996 od for field camp 3.1 ield, climbing, and camping gear 1.7 Commodities Total \$4.8
1 SCA volunteer in Homer, 3 months 3.4 cleaning and repair of radios and outboard motors 0.6 When a non-trustee organization is used, the form 4A is required. Contractual Total Commodities Costs: Proposed Description FFY 1996 odd for field camp 3.1 ield, climbing, and camping gear 1.7 Commodities Total \$4.8
Cleaning and repair of radios and outboard motors 0.6 When a non-trustee organization is used, the form 4A is required. Contractual Total \$10.6 Commodities Costs: Proposed Description FFY 1996 ood for field camp 3.1 ield, climbing, and camping gear 1.7 Commodities Total \$4.8
When a non-trustee organization is used, the form 4A is required. Contractual Total \$10.6 Commodities Costs: Proposed Description FFY 1996 ood for field camp 3.1 ield, climbing, and camping gear 1.7 Commodities Total
When a non-trustee organization is used, the form 4A is required. Contractual Total \$10.6 Commodities Costs: Proposed Description FFY 1996 ood for field camp 3.1 ield, climbing, and camping gear 1.7 Commodities Total
When a non-trustee organization is used, the form 4A is required. Contractual Total \$10.6 Commodities Costs: Proposed Description FFY 1996 ood for field camp 3.1 ield, climbing, and camping gear 1.7 Commodities Total S4.8
When a non-trustee organization is used, the form 4A is required. Contractual Total \$10.6 Commodities Costs: Proposed Description FFY 1996 iood for field camp 3.1 ield, climbing, and camping gear 1.7 Commodities Total \$4.8
When a non-trustee organization is used, the form 4A is required. Contractual Total \$10.6 Commodities Costs: Proposed Description FFY 1996 iood for field camp 3.1 'ield, climbing, and camping gear 1.7 Commodities Total S4.8
When a non-trustee organization is used, the form 4A is required. Contractual Total \$10.6 Commodities Costs: Proposed Description FFY 1996 ield, climbing, and camping gear 3.1 1.7 1.7 Commodities Total \$4.8
When a non-trustee organization is used, the form 4A is required. Contractual Total \$10.6 Commodities Costs: Proposed Description FFY 1996 iood for field camp 3.1 iield, climbing, and camping gear 1.7 Commodities Total \$4.8
When a non-trustee organization is used, the form 4A is required. Contractual Total \$10.6 Commodities Costs: Proposed Description FFY 1996 ood for field camp 3.1 ield, climbing, and camping gear 1.7 Commodities Total
Commodities Costs: Proposed Description FFY 1996 iood for field camp 3.1 ield, climbing, and camping gear 1.7
Description FFY 1996 food for field camp field, climbing, and camping gear 1.7
iood for field camp 3.1 field, climbing, and camping gear 1.7 Commodities Total \$4.8 FORM 3B FORM 3B
field, climbing, and camping gear 1.7
Commodities Total \$4.8
FORM 3B
Project Number: 96163J
AOOC Project Title: APEX/Barren Islands Murres and Contractua
1330 Kittiwakes
Commoditie
s

October 1, 1995 - September 30, 1996

New Equipment Purchases:	umber	Unit	Proposed
Description a	f Units	Price	FFY 1996
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
Those purchases associated with replacement equipment should be indicated by placement of an R. Ne	w Equ	ipment Total	\$0.0
Existing Equipment Usage:	1	Number	Inventory
Description		of Units	Agency
· · ·			
Broject Number: 961621			
Project Title: APEV/Perron Talanda Murros and	I	۲_۲	OKM 3B
1996 Rittiuskog		Eq	uipment
Adency: DOI	ļ		DETAIL
Agency. Dor		L	

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	Authorized	Proposed						and the second sec
Budget Category:	FFY 1995	FFY 1996						
Personnel	\$4.1	\$8.4						
Travel	\$0.4	\$0.5						
Contractual	\$3.6	\$3.7						
Commodities	\$2.0	\$2.0						
Equipment	\$0.0	\$0.0		LONG RA	NGE FUNDIN	IG REQUIREN	MENTS	
Subtotal	\$10.1	\$14.6	Estimated	Estimated	Estimated	Estimated	Estimated	Estimated
General Administration	\$0.9	\$1.5	FFY 1997	FFY 1998	FFY 1999	FFY 2000	FFY 2001	FFY 2002
Project Total	\$11.0	\$16.1	\$16.0	\$16.0	\$16.0			
Full-time Equivalents (FTE)	0.1	0.2						
			Dollar amount	s are shown ir	thousands of	dollars.		
Other Resources								
Comments: Forage fish will be	obtained from	the stomachs	s of sport caugh	nt large fish pr	redators to test	t the feasibility	and effective	ness of
obtaining low cost, spatial and r	elative abunda	ance data on f	orage fish in th	e Gulf of Alas	ska.			
					*			
								i
	r							
							[FORM 2X
	Project	Number: 9	6163K					FURM JA
1996	Project	Title: AP	PEX/Large	Fish as	Samplers			AGENCY
	Agency	DOT /USEWS	Ling Line ge	- 2011 0.0 1	Comprese			PROJECT
	ingency.	POT / OPENS	,					DETAIL
Prepared: 4/28/95	L						L	

				A A a make - I			<u> </u>
Per	sonnei Costs:		GS/Range/	Months	Monthly	• •	Proposed
PM		Position Description	Step	Budgeted	Costs	Overtime	FFY 1996
	D. Roseneau	PI	GS11/4	2.0	4,200		8.4
							0.0
							0 .0
		•				i	0.0
1							0.0
							0.0
							0.0
							0.0
							0.0
H							0.0
							0.0
							0.0
		Subtotal		2.0	4,200	0	
Tho	se costs associated with pro-	gram management should be indicated by	placement of	an *.	Per	sonnel Total	\$8.4
Tra	vel Costs:		Ticket	Round	Total	Daily	Proposed
РM	Description		Price	Trips	Days	Per Diem	FFY 1996
	Homer to Seward		275	1	1	225	0.5
							0.0
		,					0.0
							0.0
Į.	ļ						0.0
							0.0
1							0.0
1							0.0
							0.0
1							0.0
	1						0.0
1							0.0
Tho	se costs associated with pro	gram management should be indicated by	placement of	an *.		Travel Total	\$0.5
					1	<u> </u>	CODM 2D
1		Project Number: 96163K					
	1996	Project Title: APEX/Large	Fish as	Samplers	1		ersonnel
1		and a second and a	1 TOU 00 .	Jampiero	1	<u>ک</u> ا	Travel

Personnel & Travel DETAIL

Agency: DOI/USFWS

Contractual Costs:			Proposed
Description			FFY 1996
1 SCA volunteer in Homer for 3	months		3.7
When a non-trustee organizatio	n is used, the form 4A is required.	Contractual Total	\$3.7
Commodities Costs:			Proposed
Description			FFY 1996
sampling supplies and freight	· · · · · · · · · · · · · · · · · · ·		2.0
		Commodities Total	\$2.0
1996	Project Number: 96163K Project Title: APEX/Large Fish as Samplers Agency: DOI/USFWS	F Cor Con	ORM 3B htractua l & nmoditie s

New Equipment Purchases:	Number	Unit	Proposed
Description	of Units	Price	FFY 1996
			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 an R.	New Equ	ipment Total	\$0.0
Existing Equipment Usage:		Number	Inventory
Description		of Units	Agency
·			
		F	OPM 3B
AOOC Project Number: 96163K]		uinmon+
Project Title: APEX/Large Fish as Samplers			
Agency: DOI/USFWS	l		JETAIL
		L]

Authorized Proposed Budget Category: **FFY 1995 FFY 1996** \$3.7 Personnel \$3.6 Travel \$0.0 \$0.0 \$0.0 \$0.0 Contractual Commodities \$0.0 \$0.0 \$0.0 LONG RANGE FUNDING REQUIREMENTS Equipment \$0.0 \$3.6 \$3.7 Estimated Estimated Estimated Estimated Subtotal Estimated Estimated \$0.5 FFY 1997 FFY 1998 FFY 1999 **FFY 2000** General Administration \$0.6 **FFY 2001** FFY 2002 \$4.1 \$4.3 \$5.0 \$5.0 \$5.0 **Project Total** Full-time Equivalents (FTE) 0.3 0.3 Dollar amounts are shown in thousands of dollars. Other Resources Comments: Forage fish will be obtained from the stomachs of sport caught large fish predators to test the feasibility and effectiveness of obtaining low cost, spatial and relative abundance data on forage fish in the Gulf of Alaska. FORM 3A Project Number: 96163K AGENCY 1996 Project Title: APEX/Large Fish as Samplers PROJECT Agency: DOI/NPS DETAIL Prepared: 4/28/95

4/28/95

October 1, 1995 - September 30, 1996

Per	sonnel Costs:		GS/Range/	Months	Monthly		Proposed
PM	Name	Position Description	Step	Budgeted	Costs	Overtime	FFY 1996
		NPS volunteer		4.0	925		3.7
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
 		Subtot	al	4 0	925	0	
Tho	se costs associated with	program management should be indicated t	by placement of	an *.	Per	sonnel Total	\$3.7
Tra	/el Costs:		Ticket	Round	Total	Daily	Proposed
PM	Description		Price	Trips	Days	Per Diem	FFY 1996
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
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							0.0
l							0.0
Tho	se costs associated with	program management should be indicated t	I of the second of the second	<u>I l</u>		Travel Total	<u>5.0</u>
L			y placement of	۳			
							
		Project Number: 96163K					UKM 3B

1996

Project Number: 96163K Project Title: APEX/Large Fish as Samplers Agency: DOI/NPS FORM 3B Personnel & Travel DETAIL

Contractual Costs: Proposed FFY 1996 Description \$0.0 **Contractual Total** When a non-trustee organization is used, the form 4A is required. Proposed Commodities Costs: **FFY 1996** Description • **Commodities Total** \$0.0 FORM 3B Project Number: 96163K Contractua 1996 Project Title: APEX/Large Fish as Samplers 1 & Agency: DOI/NPS Commoditie

New Equipment Purch	hases:	Number	Unit	Proposed				
Description		of Units	Price	FFY 1996				
				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 assoc	ciated with replacement equipment should be indicated by placement of an R.	New Equ	ipment Total	\$0.0				
Existing Equipment Usage:			Number	Inventory				
Description		of Units	Agency					
1996	Project Number: 96163K Project Title: APEX/Large Fish as Samplers Agency: DOI/NPS		F Eq 1	ORM 3B uipment DETAIL				
	Authorized	Proposed		and the second	u alara a a a guter a a g	and an all the state of the sta		
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Budget Category:	FFY 1995	FFY 1996						
Dudget outogory.								
Personnel	\$8.0	\$20.0						
Travel	\$1.2	\$3.1						
Contractual	\$10.0	\$10.0						
Commodities	\$5.0	\$5.0						
Equipment	\$2.5	\$2.5		LONG RA	NGE FUNDIN	IG REQUIRE	MENTS	
Subtotal	\$26.7	\$40.6	Estimated	Estimated	Estimated	Estimated	Estimated	Estimated
General Administration	\$1.9	\$3.7	FFY 1997	FFY 1998	FFY 1999	FFY 2000	FFY 2001	FFY 2002
Project Total	\$28.6	\$44.3	\$44.0	\$44.0	\$44.0			
Full-time Equivalents (FTE)	0.3	0.3						
			Dollar amounts	s are shown ir	1 thousands of	dollars.		
Other Resources			T T		l l			
Comments: The primary functi	ion of this como	onent will be t	lo expand the l	Minerals Mana	agement Servi	ce's Cook Inte	UGulf of Alas	a forage
fish study. This component will	l also coordinate	the continua	tion of the hist	nric review of	the ecosystem	structure in t	he Prince Will	iam
Sound/Gulf of Alaska complex	Included in thi	s review will h	e obtaining an	d synthesizing	n several foran	e fish data se	te	
			o obtaining an	o synthesizing	g Several lotag		(3.	
L				an 11. An				
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	Project 1	Number: 9	6163L					FORM 3A
4000	Project 1	Title: AP	EX/Barren	s Is, Su	rvev & Hi	storic		AGENCY
1996	Review		Lif Bur z Ch	- 101 00	riel a ur			PROJECT
	Adency -					1	'	DETATI.
	Ingency. L	501						
Prepared: 4/28/95	L							

Per	sonnel Costs	:		GS/Range/	Months	Monthly		Proposed
PM	Name		Position Description	Step	Budgeted	Costs	Overtime	FFY 1996
	J. Piatt		PI	3	4.0	5,000		20.0
								0.0
								0.0
								0.0
								0.0
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l								0.0
			Sublatal		4.0	5 000		0.0
Tho	se costs asso	ciated with prov	aram management should be indicated by	placement of	an *	Per	sonnel Total	\$20.0
Tra	el Costs:			Ticket	Round	Total	Daily	Proposed
PM	Description			Price	Trips	Davs	Per Diem	FFY 1996
<u> </u>	Anchorage to	Seattle		1112	2	4	225	3.1
1								0.0
								0.0
								0.0
								0.0
								0.0
1								0.0
1								0.0
								0.0
								0.0
								0.0
			erem management of suid he indicated he				Traval Tatal	0.0
	se cosis asso	ciated with pro-	gram management should be indicated by	placement of		<u></u>	Traver Total	J
r			ſ		· · · · · · · · · · · · · · · · · · ·			
1			Project Number: 96163L					FORM 3B
1	1006		Project Title: APEX/Barren	ns Is. Su	rvey & Hi	storic	P	ersonnel
	1330		Review		-		3	Travel
	[Agency: DOI					DETAIL

1996 EXXON VALDEZ TRUSILE COUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

Contractual Costs		T	Proposed
Description			FEV 1006
Barren Islands vessel sur	νεν		10.0
	ningting is used, the form (A) is required	Total	£10.0
when a non-trustee organ	nization is used, the form 4A is required.	ractual Iotal	\$10.0 Decenced
Commodifies Costs:			Proposed
vessel fuel			50
			0.0
		dial - Total	<u> </u>
	Commo	onties rotal	<u>ا ۵.c4</u>
<u> </u>		E	OPM 3P
	Project Number: 96163L		
1996	Project Title: APEX/Barrens Is. Survey & Historic		
	Review		
	Agency: DOI	Con	mourcre

New Equipment Purchases:	Number	Unit	Proposed
Description	of Units	Price	FFY 1996
Misc. computer software and hardware			2.5
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
		Total	0.0
Those purchases associated with replacement equipment should be indicated by placement o	an R. New Equ	ipment iotai	\$2.5
Existing Equipment Usage:		Number	Inventory
			Agency
			
		······································	
Project Number: 961631			
Droject mitle: ADEV/Barrong To Survey	L Historia		OKW 3R
1996 Poviou		Eq	uipment
Review DOI			DETAIL
Agency: DOI		L	
	5		

	Authorized	Proposed		eren er er er	a character and the second	esta pre procesaria e a	s versék élek élekter	
Budget Category:	FFY 1995	FFY 1996						
Personnel	\$6.2	\$8.4						
Travel	\$0.0	\$0.0						
Contractual	\$0.0	\$0.0						
Commodities	\$0.0	\$0.0						
Equipment	\$0.0	\$0.0		LONG RA	NGE FUNDIN	IG REQUIRE	MENTS	
Subtotal	\$6.2	\$8.4	Estimated	Estimated	Estimated	Estimated	Estimated	Estimated
General Administration	\$0.9	\$1.3	FFY 1997	FFY 1998	FFY 1999	FFY 2000	FFY 2001	FFY 2002
Project Total	\$7.1	\$9.7	\$10.0	\$10.0	\$10.0			
		· · · · · · · · · · · · · · · · · · ·						
Full-time Equivalents (FTE)	0.3	0.3						
			Dollar amount	s are shown in	thousands of	dollars.		
Other Resources								
Comments' This component	will continue the	historic review	v of the ecosy	stem structure	in the Prince	William Soun	d/Gulf of Alask	a complex
			-					

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Pers	sonnel Costs):		GS/Range/	Months	Monthly		Proposed
PM	Name		Position Description	Step	Budgeted	Costs	Overtime	FFY 1996
			biologist	GS9	4.0	2,100		8.4
								0.0
								0.0
								0.0
								0.0
								0.0
								0.0
								0.0
								0.0
								0.0
								0.0
┠───┘			Cublete		4.0	2 100		0.0
The		ciated with pro	Subiolal	olacement of	4.0	2,100	U connel Total	A 83
TTTO:		clated with pro	gram management should be indicated by	Tickot	an . Dound	Total	Doily	Proposed
Dia	Deservation			Brice		Totar	Dally Der Diem	EEV 1006
	Description			r nee	inps	Days	Fei Diein	0.0
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			,		· · ·			0.0
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								0.0
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Ino	se costs asso	ciated with pro	gram management should be indicated by	placement of	an ".		Travel Total	\$0.0
		1	ſ <u></u>				r	
			Project Number: 96163L					FORM 3B
	1006		Project Title: APEX/Histor	ric Revie	w of Forad	ge Fish	P	ersonnel
1	1330		Data			-	8	Travel
			Agency: NOAA					DETAIL

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Contractual Costs:			Proposed
Description			FFY 1996
When a non-trustee organizatio	n is used, the form 4A is required. Contra	actual Total	\$0.0
Commodities Costs:			Proposed
Description			FFY 1996
	· Commo	dities Total	\$0.0
1996	Project Number: 96163L Project Title: APEX/Historic Review of Forage Fish Data Agency: NOAA	F Cor Con	ORM 3B htractua l & nmoditie S

New Equi	pment Purchases:	Number	Unit	Proposed
Descriptio	n	of Units	Price	FFY 1996
				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 pur	chases associated with replacement equipment should be indicated by placement of an R	. New Equ	ipment Total	\$0.0
Existing I	Equipment Usage:		Number	Inventory
Descriptio	n		of Units	Agency
		* *		
		i		
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· ····				
	Project Number: 96163L		F	ORM 3B
100	Project Title: APEX/Historic Review of Fo	rage Fish	Ea	uipment
132	Data			DETAIL
	Agency: NOAA			

Budget Category	Authorized	Proposed	Sector references and					이 이 이 이 이 이 이 같은 한 것을 했다.
IDUQUEL CALEGORY:	FFY 1995	FFY 1996						
Personnel	\$16.6	\$16.8						
Travel	\$0.0	\$0.0						
Contractual	\$0.0	\$0.0	ŝ					
Commodities	\$0.0	\$0 .0						
Equipment	\$0.0	\$0.0		LONG RA	NGE FUNDIN	G REQUIREN	AENTS	
Subtotal	\$16.6	\$16.8	Estimated	Estimated	Estimated	Estimated	Estimated	Estimated
General Administration	\$2.5	\$2.5	FFY 1997	FFY 1998	FFY 1999	FFY 2000	FFY 2001	FFY 2002
Project Total	\$19.1	\$19.3	\$19.0	\$19.0	\$19.0			
				en este Branzelle en	an an gara tanan 10 Milang baga sana Milang sana sana sana sana sana sana sana s			
Full-time Equivalents (FTE)	0.6	0.6						
			Dollar amounts	s are shown in	thousands of	dollars.		
Other Resources								
					۰.			
	9							

1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

Per	sonnel Costs	•		GS/Range/	Months	Monthly		Proposed
PM	Name		Position Description	Step	Budgeted	Costs	Overtime	FFY 1996
			biologist (Homer)	16	3.0	2,400		7.2
ĥ			biologist (Kodiak)	16	4.0	2,400		9.6
ł						0		0.0
								0.0
1								0.0
								0.0
ļ								0.0
								0.0
1	1							0.0
]								0.0
1								0.0
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<u>ال</u>			Subtotal		7.0	4,800	0	
Tho	se costs asso	ciated with pro	gram management should be indicated by	placement of	an *	Per	sonnel Total	\$16.8
Tra	vel Costs:			Ticket	Round	Total	Daily	Proposed
PM	Description			Price	Trips	Days	Per Diem	FFY 1996
8								0.0
	ļ				· •			0.0
								0.0
	1							0.0
l I								0.0
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			grant management should be mulcated by	placement of			IIAVEL LUIAL	<u>۵</u> 0.0
<u> </u>				······································				
1			Project Number: 96163L					FORM 3B
1	1996		Project Title: APEX/Histon	ric Review	w of Fora	ge Fish	P	ersonnel
1	1330		Data				8	Travel

& Travel DETAIL

Agency: ADF&G

Contractual Costs:	Proposed
Description	FFY 1996
When a non-trustee organization is used, the form 4A is required. Contractual	Fotal \$0.0
Commodities Costs:	Proposed
Description	FFY 1996
Commodities 1	otal \$0.0
1996 Project Number: 96163L Project Title: APEX/Historic Review of Forage Fish Data Agency: ADF&G	FORM 3B Contractua l & Commoditie

1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

New Equipment Purchases:	Number	Unit	Proposed
Description	of Units	Price	FFY 1996
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
			0.0
	{		0.0
			0.0
Those purchases associated with replacement equipment should be indicated by placement of an R	New Fau	ioment Total	<u> </u>
Frieding Equipment [[sage:	Hen Equ	Number	
Existing Equipment Usage.		of Units	Anency
		01 01113	/\geney
			l
Project Number: 961631			
Project Title: APEX/Historic Peview of Fora	ge Fish	1	UKM JB
1996 Data	9~ 1 1011	Eq	uipment
			JETALL
Agency. Aurau	ľ	L	

8/25/95 Vers N Dritevinn Buchsets for 96076 961918 96163A-L

August 2, 1995

MEMORANDUM TO:	Traci Cramer Director of Administration
FROM:	Bruce Wright Program Manager
Re:	Interim Budgets (Oct. 1, 1995 to Jan. 31, 1996)

Restoration Study 96076: Effects of Oil on Straying and Survival

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

1) <u>Maintenance and monitoring of incubation system</u>. Daily monitoring of flow rates to 100 incubators, cleaning of incubators as necessary, and monitoring and maintenance of seawater and freshwater supply systems.

2) <u>Evaluation of treatment effects</u>. Assessment of survival of embryos to eyed-stage (late October-early November) and to hatching (January).

3) <u>Hydrocarbon analysis</u>. Collection of gravel, water, and embryo samples at eyed stage and hatch stage, and analysis of samples collected at earlier spawning in FY 95.

4) <u>Sashin Creek weir operation</u>. Continued operation of Sashin Creek weir until late October to enumerate total escapement of pink salmon to Sashin Creek. This requires daily fish counts and weir cleaning.

5). Stream Surveys. Weekly carcass surveys of Sashin and Lovers Cove Creeks in October

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

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

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

COSTS BY LINE ITEM, 96076

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

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

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

COSTS BY LINE ITEM, 96191B

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

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

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

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

COST	COSTS BY AGENCY and BY LINE ITEM, 96163 A-L										
Line Item	<u>DOI (\$K)</u>	<u>NOAA (\$K)</u>	<u>ADF&G (\$K)</u>	<u>TOTAL (\$K)</u>							
Personnel	181.1	29.3	26.3	, 236.7							
Travel	7.0	7.0	0.0	14.0							
Contracts	0.0	0.0	0.0	0.0							
Commodities	0.0	0.0	0.0	0.0							
Equipment	0.0	0.0	0.0	0.0							
SUBTOTAL	188.1	36.3	26.3	250.7							
General Admin	27.2	4.4	3.9	35.5							
TOTAL	\$ 215.3	\$ 40.7	\$ 30.2	\$ 286.2							

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

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PROJECT	TITLE	Personnel Costs (title/months)	Travel Costs	Total
95163 A	Fish Survey & Biology	\$ 5.8K (PM NOAA/1 months)	\$ 1.0K	\$ 6.8K
95163 B	Bird/Fish Interactions	19.2 (PI/4 months) 5.5 (PM DOI/1 month)	0.5 0.0	25.2
95163 C	Fish Diet Overlap	9.9 (PI/3 months) 20.3 (techs./3 months) 5.8 (PM NOAA/1 month) 1.2 (PM ADFG/.2 month)	4.5 0.0 0.0 0.0	41.7
95163 D	Puffins as Samplers	5.0 (PI/1 month) 6.0 (Assis. PI/3 months)	1.0 0.0	12.0
95163 E	Black-legged Kittiwakes	19.2 (PI/4 months) 4.9 (2 techs./2 months) 5.5 (PM DOI/1 month)	1.0 0.0 0.0	30.6
95163 F	Pigeon Guillemots	19.2 (PI/4 months) 4.9 (2 techs./2 months) 5.5 (PM DOI/1 month)	1.0 0.0 0.0	30.6
95163 G	Energetics	2.8 (PM NOAA/0.5 month)	1.0	3.8
95163 H	Proximate Composition	0.0	0.0	0.0
95163 I	Project Leader	35.0 (PI/2 months) 2.5 (PM DOI/0.5 month) 2.9 (PM NOAA/0.5 month) 15.0 (Fish Symposium)	1.0 0.0 0.5 0.0	56.9
95163 J	Barren I. Murres & BLKs	12.6 (PI/3 months) 6.9 (bio. tech./2 months)	1.0 0.0	20.5
95163 K	Fish as Samplers	4.2 (Pl/I month)	0.5	4.7
95163 L	Barrens & Historical	10.0 (P1/2 months) 2.1 (biologist NOAA/1 mo) 4.8 (biologists F&G/2 mo)	1.0 0.0 0.0	17.9
General Admin. TOTAL		236.7*	14.00	250.7*

96	163	A-L	Interim	Budget	for	Oct.	1.	1995	to	Jan.	31.	1996

PI = principal investigator PM = program manager * General administration will be added to these costs. See summary tables below.

PACIFIC HERRING PROJECTS COORDINATION

Project Number:	96164
Restoration Category:	Research
Proposer:	ADF&G
Lead Agency:	ADF&G
Cooperating Agencies:	UAF, NOAA, PWSSC
Duration:	4 years
Cost FY 96:	\$49,200
Cost FY 97:	\$49,200
Cost FY 98:	\$49,200
Cost FY 99:	\$49,200
Cost FY 00:	
Cost FY 01:	
Cost FY 02:	
Geographic Area:	Prince William Sound
Injured Resource/Service:	Pacific Herring/Commercial fishing/ Subsistence

ABSTRACT

The purpose of this project will be to enhance coordination, integration and critical review of projects that are designed to study different aspects of Pacific herring in the Prince William Sound ecosystem; to better understand the interactions of the components of the ecosystem; and, to aid in the recovery of the injured resource and lost services.

INTRODUCTION

Pacific herring, Clupea harengus pallasi, play a vital role in the ecosystem and economy of Prince

William Sound (PWS). Within the ecosystem, they occupy the middle link of the food chain as they provide an energy source for many other species as forage for other fish, birds, sea mammals and invertebrates. Since the *Exxon Valdez* oil spill, this link was broken and the Pacific herring populations are depressed. Consequently, a number of research and monitoring projects were proposed and funded by the Trustee Council (TC) in an attempt to better understand the dynamics of the Pacific herring in PWS and to aid the restoration of the Pacific herring populations.

As the number and complexity of the Pacific herring studies expanded, it became apparent that there is a need to assure that there is good communication between the Principal Investigators (PIs) within the Pacific herring studies and between these PIs and PIs of other study programs. The purpose of this project is to provide greater organization and coordination among the projects that study Pacific herring and their role in the PWS ecosystem. The scope of this project will also include a broader perspective to assure good communications between PIs for Pacific herring projects and the PIs of other PWS ecosystem projects (e.g. APEX; Nearshore Predators; SEA). It will employ the services of recognized fisheries experts who specialize in herring biology to incorporate their overview, review and comments from an external perspective into hypotheses, reports and proposals.

NEED FOR THE PROJECT

A. Statement of Problem

Pacific herring, *Clupea harengus pallasi*, has been identified by the TC as a resource that was injured but has not recovered. The oil spill occurred as the Pacific herring were spawning in 1989. Studies of injuries to adult Pacific herring began immediately and histopathological damage was measured in adults in 1989 and 1990. Over 40% of the PWS spawning areas were oiled. Physical and genetic abnormalities in newly hatched Pacific herring larvae and reduced hatching success were observed.

The spawning populations in 1993, 1994 and 1995 have been much below expectations for PWS. Pathological studies have indicated an elevated incidence of the pathogens, viral hemorrhagic septicemia (VHSV) and *Ichthyophonus*. These have been implicated in the cause of the mortality and possible suppression of the recovery but it is unknown if the disease incidence is in response to oil exposure in 1989 and 1990 or if it is in response to other disturbances to the ecosystem.

B. Rationale

Since the oil spill and the crash of the Pacific herring populations in PWS, there has been considerable interest in Pacific herring damage assessment and restoration. A Draft "Stock Model for Pacific Herring in Prince William Sound" was prepared by Brown and Wilckck (1994) to propose a hypothetical stock structure and a detailed description of the assumptions upon which it was based. Studies funded by the TC have attempted to assess the role of Pacific herring in the PWS ecosystem, the impacts of the ecosystem on Pacific herring and the enhancement of management studies to monitor Pacific herring populations and the recovery status (Table 1). Restoration strategies will depend on understanding the cause of the 1993 crash, the role of interactions within the ecosystem and the best available stock status information. Each Pacific herring project has different objectives and the PIs for these projects have diverse backgrounds and expertise and they work for different

agencies or organizations. Some projects are components of other TC-funded programs such as the Prince William Sound Ecosystem Assessment (SEA) Program. All Pacific herring projects are important to other components of the Prince William Sound ecosystem as well because Pacific herring are a vital link of the food web that may affect populations of both producers (e.g. zooplankton) and consumers (e.g. marine mammals, birds and fish predators). All are affected by the primary productivity of the ecosystem, which, in turn, is driven by oceanic currents. Some of these complex interactions are indicated in Figure 1. Consequently, it is imperative that Pacific herring project PIs communicate well with each other and with PIs of other projects and that project activities are well coordinated.

This project is intended to enhance communication and coordination among Pacific herring PIs and with PIs of other projects that study resources that are interrelated and affected by the recovery of Pacific herring. This will assure that projects and tasks within projects will be complementary and not overlapping, and that information derived from Pacific herring projects will be shared with PIs of other projects and that data that they need will be collected. Extra quality assurance will be provided by inviting constructive criticism from experts outside the oil spill process.

C. Summary of Major Hypotheses and Objectives

The purpose of this suite of projects is to determine and model the interactions of Pacific herring with other components within the PWS ecosystem to understand the impacts of depressed Pacific herring stocks and approaches that may aid the restoration of this injured species. Objectives addressed by the various projects (specific objectives are listed within each DPD of each project) include:

- Spawning biomass and spawning deposition
- Mortality factors and survival rate of Pacific herring eggs
- Role of disease in population control
- Juvenile growth, habitats and migration
- Energy cycles for overwintering survival and reproductive success
- Stock identification and management applications
- Role of oil in long-term damage
- Assessment of Pacific herring reproductive impairment status

D. Completion Date

The overall goal of this project will be met when the results and objectives from each of the individual projects have been met (Table 1) and incorporated into the SEA Model and conclusions are reported in a final synopsis review report. These conclusions will also be shared for incorporation into summary reports of other programs and projects that describe the components of

the PWS ecosystem. This overall goal is expected to be completed in FY 1999.

COMMUNITY INVOLVEMENT

Individual projects within the Pacific herring study group and within the SEA program will have their own plan to involve residents from the spill area communities and to incorporate local/traditional knowledge. Obviously, there is strong interest in Pacific herring population recovery status and the overall recovery of the ecosystem in Prince William Sound. Hypotheses, results and conclusions derived from these projects will be communicated to the residents indirectly through the TC and directly by individual PIs.

FY96 BUDGET

14.9
10.7
20.0
0.0
0.0
45.6
3.6
49.2

Project No.	Project Title	Principal Investigator	Completion
95074	074 Reproductive Impairment		1997
95165	Genetic Structure	J. Seeb	1998
95166	Herring Natal Habitat	Willette	1998 ¹
95320E	Juvenile Salmon and Herring Integration	Willette	1998
95320N	Sound Ecosystem Assessment (SEA) Nekton-Plankton Acoustics	Thomas	1998
953208	Investigations of Disease Factors Affecting Declines of Pacific Herring Populations in Prince William Sound, Alaska	Kocan	1998
95320T	Juvenile Herring Growth and Habitat Partitioning	Norcross	1998
95320U	Somatic and Spawning Energetics of Herring and Pollock	Paul	1998

Table 1. Exxon Valdez Trustee Council Pacific Herring Projects, 1996.

¹ Major components will be completed sooner.

PWS Herring Research in the Near Future



Figure 1. Pacific herring research projects in Prince William Sound

PROJECT DESIGN

A. Objectives

- 1. Coordination and integration of hypotheses, objectives, tasks, schedules, data collections and conclusions among PIs within the Pacific herring studies.
- 2. Coordination and integration of hypotheses, objectives, data needs and conclusions among PIs of other PWS ecosystem research projects.
- 3. Integration of critical evaluation among the Pacific herring study group PIs by specialists in the field of herring biology who are presently not associated with the TC research.
- 4. Prepare a final synopsis review report that integrates information from all of the Pacific herring studies and the other PWS ecosystem studies.

B. Methods

- 1. Coordination within Pacific herring studies.
 - a. A discussion, network or forum will be established by the Program Manager among the PIs to enhance information exchange. PIs will be requested to provide brief (e.g., one page) summaries of activities whenever a significant event is completed (e.g., a spawning event or a data collection trip) to share important findings or observations.
 - b. Quarterly reports will be assembled as a Pacific herring package and distributed among the researchers.
 - c. An annual meeting of the TC Pacific herring project PIs will be convened in early tail to discuss annual findings and compare results and draft conclusions.
 - d. During the Annual Workshop, a special session will be convened for the TC Pacific herring project PIs to review results and conclusions, proposals and hypotheses for the next FY, data gaps, work schedules, common tasks and special needs.
- 2. <u>Coordination and integration with other ecosystem research studies</u> will occur with presentations and participation during the Annual Restoration Workshop and invitations to other PIs to participate in the Pacific herring special session.

3. Integration of critical evaluation

- a. Two specialists in the field of herring biology will be contracted to review and provide critical comments on project proposals and project reports.
- b. The specialists will participate in the Pacific herring study review meeting and the Annual Restoration Workshop.

- c. The herring specialists will be selected to assure that they have different backgrounds to provide a broad scope of coverage to accommodate the broad range of topics and review needs of the diverse Pacific herring projects. Selection Criteria will include: publication record, specialty area, reputation as a scientist, written and oral communication ability, committment and availability.
- 4. <u>Final synopsis report</u>. One of the herring specialists, the Program Manager or one of the Pacific herring PIs will be designated to write a final synopsis report to summarize and integrate the results and conclusions from all of the Pacific herring studies.
- 5. <u>Communication</u> The Program Manager for the Alaska Department of Fish and Game (ADF&G) will assure that the means of communication for integration, coordination and review will be established and maintained and that contracts are administered. The Program Manager will be responsible to prepare the agenda and make arrangements for the Pacific herring workshops.

C. Contracts and Other Agency Assistance

Contracts will be required for two specialists in herring biology. Specific tasks will include:

•Review and comments for Pacific herring project proposals.

•Review and comments for Pacific herring draft progress reports.

•Travel and per diem to participate in a Pacific herring review meeting and the Annual Restoration Workshop.

It is expected that these tasks can be accomplished in approximately one month annually. This will allow one week, twice yearly, to review materials and prepare for the workshops and one week, twice yearly, to participate in the workshops.

D. Location

The Pacific herring projects are focused within Prince William Sound. Collectively, they will provide critical information to help to understand linkages between the components of the Prince William Sound ecosystem.

SCHEDULE

A. Measurable Project Tasks for FY96

- Start-up to November 15: Advertise and contract herring specialists
- By December 15: Convene annual Pacific herring fall workshop to review results and draft conclusions

January/February: Convene special session of the Annual Restoration Workshop to review results and conclusions, proposals for FY 1997 sampling schedules and data needs.

B. Project Milestones and Endpoints

- 1. The objectives of coordination and integration of hypotheses, conclusions, work schedules and data collections as well as review and critical comment for project proposals and project reports will be accomplished each year.
- 2. The objective of preparing a final synopsis review report that is expected to be met at the end of the period of study for these projects in 1999.

C. Project Reports

Each Pacific herring project will remain as a separate entity and each will have a schedule for annual and final project reports. This project will, however, is expected to provide a final synopsis review report in FY1999.

COORDINATION AND INTEGRATION OF RESTORATION EFFORT

The purpose of this project is to create a means for coordination and integration with other restoration efforts and the SEA model. The primary intent is to increase efficiency, information exchange and collective expertise among the PIs within the Pacific herring projects group. The secondary intent is to assure that this increased expertise and the knowledge about Pacific herring in Prince William Sound is distributed and shared among all ecosystem study projects in Prince William Sound. Finally, results and conclusions from these studies will be communicated to individuals, communities and agencies outside the TC.

It will be the responsibility of the Program Manager for ADF&G to assure that the lines of communication are established and maintained and contracts are administered.

ENVIRONMENTAL COMPLIANCE

Compliance with the National Environmental Policy Act (NEPA) will be required for this project. This will most likely result in a decision as a Categorical Exclusion (CE) and the most likely lead federal agency will be the National Oceanographic and Atmospheric Administration (NOAA).

PERSONNEL

William J.Hauser:

Education-

B.S., Zoology, University of Wisconsin, 1965

M.S., Fish and Wildlife Management, Montana State University, 1968 Ph.D., Zoology, University of Maine, 1973

Professional Experience-

1994-present:	Assistant Program Manager, H&R Div., ADF&G
1994:	Writing Team Member for fisheries, EVOS Restoration Plan EIS
1993-1994:	Regional Resource Development Biologist, CFMD Div., ADF&G
1980-1993:	Regional Biologist, FRED Div., ADF&G
1977-1980:	Chief Fisheries Scientist, Nalco/Hazelton Environmental Sciences
1973-1977:	Research Biologist, University of California, Riverside

Name of proposed Project Leader William J. Hauser Alaska Dept. of Fish & Game Habitat and Restoration Division 333 Raspberry Rd. Anchorage, AK 99508 Ph: (1-907) 267-2172 Fax: (1-907) 522-3148 EMail:

Name of proposed Project Manager Joseph R. Sullivan Alaska Dept. of Fish & Game Habitat and Restoration Division 333 Raspberry Rd. Anchorage, AK 99508 Tel: (1-907) 267-2213 Fax: (1-907) 522-3148 E-Mail:

Date prepared

Budget Category:	Authorized	Proposed FEV 1996						
Budget Category.	1111000	1111330						
Personnel		\$31.6						
Travel		\$5.0						
Contractual ·		\$3.1						
Commodities		\$0.5						
Equipment		\$4.0		LONG I	RANGE FUNDIN	G REQUIREME	NTS	
Subtotal	\$0.0	\$44.2	Estimated	Estimated	Estimated	Estimated	Estimated	Estimated
General Administration		\$5.0	FFY 1997	FFY 1998	FFY 1999	FFY 2000	FFY 2001	FFY 2002
Project Total	\$0.0	\$49.2						
Full-time Equivalents (FTE)		0.5						
·			Dollar amount	ts are shown in	thousands of o	dollars.		
Other Resources						l	<u> </u>	
Note: This budget accounts direction of funding and/or	tasks of the individual F	or a full-time " Pacific herring p	Pacific Herring projects.	Program Coor	dinator". The t	alance of the f	unding will be j	provided by re
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Note: This budget accounts	tasks of the individual F	or a full-time "	Pacific Herring projects.	Program Coor	dinator". The t	palance of the f	unding will be i	provided by re
Note: This budget accounts direction of funding and/or 1996	Project Numb Project Title: Agency: AK	per: 96164 Pacific Herring poer: 96164 Pacific Herri Dept. of Fisl	ng Program h & Game	Coordinator	dinator". The t	palance of the f	unding will be i	FORM 3A AGENCY PROJECT DETAIL

Contraction of the							
Pers	onnel Costs:		GS/Range/	Months	Monthly		Proposed
PM	Name	Position Description	Step	Budgeted	Costs	Overtime	FFY 1996
		Pacific Herring Program Coordinator	20A	6.0	5,265		31.6
1 2		Abore 1					0.0
							0.0
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							0.0
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							0.0
L		Subtotal		6.0	5,265	<u>,</u> 0	
Tho	se costs associated with progr	am management should be indicated by place	ment of an *.			Personnel Total	\$31.6
Trav	rel Costs:		Ticket	Round	Total	Daily	Proposed
PM	Description		Price	Irips	Days	Per Diem	FFY 1996
1	Anchorage - (Annual Rest Sci	Workshop, Herring Review)	444	2	6	125	1.6
	Anchorage		444	2	4	125	1.4
	Cordova		352	3	0	125	1.8
							0.0
							0.0
	Car Pontal \$12/day x 20		10	20			0.0
1			10	20			0.2
							0.0
1							0.0
1							0.0
							0.0
Tho	se costs associated with progr	ram management should be indicated by place	ment of an *.			Travel Total	\$5.0
							FORM 3B
		Project Number: 96164					Demonsel
	1996	Project Title: Pacific Herring Program	n Coordinato	r			Personnei
		Agency: AK Dent of Fish & Game		-		,	& Travel
		A Dept. of Fish & Galile					DETAIL
	2 of 4	<i></i>				.	8/1/95

1996 EXXON VALDEZ TRUS'1 COUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

Contractual Costs:	Proposed
Description	FFY 1996
Description Air charters -5 hrs @ \$250/hr Telecommunications, postage, and courier service WAN (CFMD network) Copying and Printing	FFY 1996 1.3 1.0 0.5 0.3
When a non-trustee organization is used, the form 4A is required.	\$3.1
Commodities Costs:	Proposed
Description	FFY 1996
Office/photo supplies	0.2
Commodities Total	\$0.5
1996 Project Number: 96164 Project Title: Pacific Herring Program Coordinator Agency: AK Dept. of Fish & Game	ORM 3B htractual & mmodities DETAIL

1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

New Equipment Pu	rchases:	Number	Unit	Proposed
Description		of Units	Price	FFY 1996
Computer		1	2,500	2.5
Printer		1	1,500	1.5
	• · · · ·			0.0
	•			0.0
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
Those purchases as	ssociated with replacement equipment should be indicated by placement of an R.	New Ed	uipment Total	\$4.0
Existing Equipment	Usage:		Number	Inventory
Deseription			of Units	Agency
				-8
				:
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	Project Number: 96164			
1996	Project Title:			quipment
	Agency: AK Dept. of Fish & Game			DETAIL
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96165

Genetic Discrimination of Prince William Sound Herring Populations 4-27 430pm

Project Number:	96165
Restoration Category	General Restoration
Proposer:	Alaska Department of Fish and Game
Lead Trustee Agency:	Alaska Department of Fish and Game
Cooperating Agencies:	None
Duration:	2 1/2 years
Cost FY 96:	105.8K
Cost FY 97:	120.0K
Cost FY 98:	97.0K
Geographic Area:	Prince William Sound
Injured Resource/Service:	Pacific herring

ABSTRACT

The Prince William Sound herring fishery has been in catastrophic decline since 1992. The Alaska Department of Fish and Game recovery effort includes incorporating a knowledge of genetically derived population structure into harvest management. In this continuing project we are delineating the structure of Prince William Sound population(s) and related North Pacific populations using both nuclear and mitochondrial DNA analyses. Tests for temporal and spatial diversity within years and temporal stability across years will be done.

INTRODUCTION

Pacific herring *Clupea pallasi* 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 near shore staging areas. Over 40% of the herring spawning, staging, and egg deposition areas and over 90% of the documented summer 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 "dipped" in 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 near shore bays and passes.

The Prince William Sound herri g fishery has been in catastrophic decline since 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. In 1994, as in 1993, the spawning population was below preseason predictions. No recovery was evident in 1995. Aerial surveys since 1993 indicated that the population was below threshold harvest levels. 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 was so low that no commercial harvest has been permitted 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. 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 Booke 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. But, previous surveys of herring using this technique have generally revealed differentiation only over broad geographic regions (Grant 1984; Grant and Utter 1984; cf., Grant et al. 1987) or between spawning populations within the same area that are temporally isolated (Kornfield et al. 1982). Allozymes define two distinct races of Pacific herring (Asian/Bering Sea and eastern North Pacific), with further subdivision between Gult 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).

Additional techniques to study the structure of natural populations have 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, that we focus upon microsatellite markers (in nuclear DNA) as being potentially most useful markers for investigation of fine structure. In consideration of those comments, and in consideration of the fact that nuclear and mitochondrial loci evolve in response to different pressures, we propose to use 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 can also be used to estimate the population composition of non-spawning aggregations contributing to the fisheries in Prince William Sound.

NEED FOR THE PROJECT

A. Statement of Problem

The Prince William Sound herring fishery is in 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

Pacific herring is 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, 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 over harvest, damage to the resource, or economic loss to the fishing industry. This information is also needed to help interpret oil spill damage results. 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.

C. Summary of Major Hypotheses and Objectives

The major hypothesis of the Brown and Wilcock (1994) stock model that we plan to test are that (1) genetic homogeneity exists within Prince William Sound and (2) the Prince William Sound population is genetically isolated from other Gulf of Alaska populations. The working objectives of this study are to:
- 1. Screen population samples (collected from spawners) using both nuclear and mitochondrial DNA approaches. Techniques will include both RFLP analysis of mitochondrial regions amplified by polymerase chain reaction (PCR) and analysis of microsatellite loci (analysis of regions with variable number of tandem repeats, VNTR).
- 2. Evaluate the null hypothesis that a single panmictic population of herring exists in Prince William Sound. Tests will include four putative population samples from both spatial and temporal isolates within the Sound.
- 3. Evaluate the structure of Prince William Sound populations within the context of the structure of adjacent spawning aggregates (up to four), including a comparison from across the known genetic barrier of the Alaska Peninsula.
- 4. Evaluate the structure of Prince William Sound and related North Pacific populations for inter-annual stability.

D. Completion Date

The duration of this project was 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 FY 95 and reporting to be complete in FY 96.

However the start date, and thus the completion date, of this project have been elusive. The Trustee Council first made funds available during FY 94 (Project 94165). The field season that year was truncated due to the surprise run failure, inadequate samples were obtained to meet project most project objectives, and project start was deferred one year. No Trustee Council funds were spent on the project in FY 94. We are in the middle of the field season for FY 95 (Project 95165) at the writing of this proposal, 96165. Some spatial isolates from within Prince William Sound have been successfully sampled, but temporal isolates remain elusive because of the current run failure. Sampling from outside of the Sound is in progress and appears adequate. We will initiate laboratory analyses with the aid of a contractor, but we are certain that FY 95 sampling will not be complete enough to meet all four project objectives.

At least two years of complete sampling are required to confirm year-to-year 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 like 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 the end of FY 98. The cover sheet for this proposal reflects place-holder budgets for FY 97 and FY 98, and those requests will be made if annual project results and Chief Scientist recommendations so indicate.

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. The project will be moved to the Alaska Sealife Center in Seward, should the project duration overlap with the availability of that facility. Again, local hire will be used when possible, and ADFG plans to participate in all of the educational and outreach programs scheduled for the Center.

FY 96 BUDGET

Personnel	22.5
Travel	2.2
Contractual Services	72.0
Commodities	.7
Equipment	00.0
Sub-total	97.4
General Administration	8.4
Total	105.8

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 depleted 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 develop harvest strategies that will speed the recovery process. In addition, it will aid local resource users to make appropriate preseason plans based on accurate and precise herring projections.

The working objectives of this study are to:

- 1. Screen samples collected from spawning aggregates using both nuclear and mitochondrial DNA approaches. Techniques will include both RFLP analysis of mitochondrial regions amplified by polymerase chain reaction (PCR) and analysis of microsatellite loci (analysis of regions with variable number of tandem repeats, VNTR).
- 2. Evaluate the null hypothesis that a single panmictic population of herring exists in Prince William Sound. Tests will include four putative population samples from both spatial and temporal isolates within the Sound.
- 3. Evaluate the structure of Prince William Sound populations within the context of the structure of adjacent spawning aggregates (up to four), including a comparison from across the known genetic barrier of the Alaska Peninsula.

4. Evaluate the structure of Prince William Sound and related North Pacific populations for inter-annual stability.

B. I lethods

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 outgroups from the Gulf of Alaska and the Bering Sea and to include tests for inter-annual stability (cf., Kornfield et al. 1982, see below).

Field collections of spawning Pacific herring will target eight representative sites within and adjacent to Prince William Sound. The collection sites within Prince William Sound will be chosen to maximize the potential genetic differentiation among temporally and spatially isolated spawning aggregations. Tissue extracts from muscle, liver, eye, and heart will be collected and preserved in liquid nitrogen until transport to -80° C freezers for archival to facilitate the option for allozyme electrophoresis at a later time. Two years of sampling will be conducted at each site to test for inter-year stability of genetic diversity measures.

The within-Sound sampling effort will target Rocky Bay, a southcentral spawning isolate; Port Gravina, a southeast isolate; and Tatitlek Narrows, a northeast isolate. Samples will be collected from both early- and late-spawning stocks in Rocky Bay. Early- and late-spawning isolates will be collected from Port Chalmers and archived for analysis during subsequent years (if further analysis of temporal isolation is deemed appropriate). One-hundred individuals will be subsampled from each aggregation during the sampling for Trustee Council Project 95166 *Herring Natal Habitat*. Consequently, age and other data will be collected from the individuals analyzed for genetic variation, facilitating further correlation analyses between population data and genetic variation.

Sampling outside of Prince William Sound will include Kodiak Island, populations thought to share an ancestral tie with Prince William Sound populations (John Wilcock, Alaska Department of Fish and Game, personal communication) and a Bering Sea population known to be genetically isolated from the other Gulf of Alaska stocks (Grant and Utter 1984). One-hundred individuals will be collected from up to four of these outgroup populations.

2. Genetic Analysis

The preproposal for this project included allozyme analysis as well as DNA analysis because allozymes have previously been shown to discriminate temporally isolated populations such as those observed in Prince William Sound (cf., Kornfield et al. 1982), and they delineate a restriction in gene flow between Bering Sea and Gulf of Alaska populations (Grant and Utter 1984). 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 that we should collect and archive samples 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). Depending upon year-one results, allozymes may be reconsidered for subsequent analyses.

Alaska Department of Fish and Game plans to seek assistance from an outside laboratory for the genetic analyses following standard State of Alaska procurement procedures for FY 95 analyses. A request for proposal will be issued for the molecular analyses to be conducted under a Reimbursable Services Agreement (RSA) with an Alaskan University or under contract from another outside laboratory. At this writing the request for proposal is pending final Trustee Council approval for Project 95165, and no analyses have been done.

Because mitochondrial and nuclear genomes evolve in response to different pressures, it is expected that the successful respondent will incorporate both approaches into the year-one screen described in this proposal. The investigator will be expected to focus upon an analysis of microsatellite loci at the recommendation of the Trustee Council's chief scientist. Details of the specific molecular techniques to be investigated will be chosen based on: 1) a review of the current literature and recently available research results, and 2) qualifications and expertise of respondents.

Alaska Department of Fish and Game plans to evaluate the option of conducting FY 96 analyses in-house (e.g., following methods described in Trustee Council Proposal 96191) after reviewing the results of project 95191 and other Department obligations.

C. Contracts and Other Agency Assistance

Alaska Department of Fish and Game laboratory staff is fully committed to other projects during the period that FY 95 analyses will be conducted. Laboratory analysis will be awarded through an RSA or through a contract awarded through the State of Alaska procurement process. FY 96 analyses may also be awarded to a contractor, depending upon an evaluation of FY 95 results and an evaluation contract efficiency.

Alaska Department of Fish and Game Genetics Laboratory has utilized the services of subcontractors on a number of Trustee Council projects. Four outside contractors were utilized for laboratory components of Trustee Council Projects 94191 and 94255. All of the laboratory analysis for Trustee Council Project 94320D was awarded on contract to a collaborating agency with a similar genetics program, Washington Department of Fish and Wildlife. Contracting some projects has increased efficiency of conducting our Trustee Council-funded research. However, costs of contracting have generally exceeded the costs of in-house research; the major exception is that graduate student research funded at universities agreeing to a 20% overhead cap has been cost-effective.

Contracting laboratory analysis will become less efficient for the department with the completion of laboratory facilities at the Alaska Sealite Center in Seward, and future contracts to outside-of-Alaska laboratories will only be awarded if the work cannot be done in the Seward facility. Some of the university subcontractors are submitting proposals to conduct projects as principal investigators (see genetics proposals to the Trustees by University of Alaska and University of Montana), and we look forward to the potential opportunity for collaboration on those projects. We propose to continue using graduate student research as an effective tool for developmental research, and we will consider

other opportunities for contracting if they present useful advantages to the State of Alaska and Trustee Council restoration program.

D. 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, archival, and data analysis will be conducted at the ADF&G area office in Cordova and regional office in Anchorage.

SCHEDULE

A. Measurable Project Tasks for FY 96

Start-up of 95165 to May 30, 1995:	Collection of samples
May 30 - June 15:	Award contract for FY 95 samples
June 15 - December 30:	Complete laboratory analysis
January - April, 1996:	Evaluate lab results, plan for 1996 sampling
April - May 30, 1996:	Collection of samples
May 30 - June 15:	Award contract or begin laboratory analysis of 1996 samples in house
June 15 - December 30:	Complete laboratory analysis
January - April, 1997:	Evaluate lab results, plan for 1997 sampling
April 1997:	Annual report for FY 96

B. Project Milestones and Endpoints

December 30, 1995:	Complete screen of population samples collected during 1994-1995
April 30, 1996:	Complete evaluation of population structure of populations collected during 1994-1995
December 30, 1996:	Complete screen of population samples collected during 1996
April 30, 1997	Evaluation of population structure of Prince William Sound and other related populations collected through 1996; planning for mop-up sample collection for spawning aggregates missed in previous years
December 30, 1997:	Complete screen of population samples collected during year three
April 30, 1998:	Complete evaluation of stability of population structure across years

C. Project Reports

April 30, 1996:	annual report in the form of manuscript submitted to journal.
April 30, 1997:	annual report.
April 30, 1998:	final report in the form of manuscript submitted to journal.

COORDINATION AND INTEGRATION OF RESTORATION EFFORT

A. Existing Agency Program

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 two months from Trustee Council funds.

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 FY 95 laboratory analyses described herein. Approximately \$50.0K of State of Alaska general funds was programmed for the study of saltwater-mediated mosaicism as the mechanism for embryo mortalities identified during implementation of Trustee Council Projects 93003 and 94191 (Miller et al. 1994). 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.

B. Other Coordination

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 96166 Herring Natal Habitats. Tissue archival and biometric analyses will be coordinated among all Trustee Council projects related to genetics including 96320D, 96191, and 96255.

Sharing of project results will be used to evaluate and revise current strategies for management of commercial herring fisheries if warranted. 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 cooperates on similar projects of conservation genetic nature on salmonids and other marine fish in the North Pacific Ocean. Alaska Department of Fish and Game is hosting the annual inter-agency meeting in Anchorage during May, 1995, where participating scientists will coordinate approaches.

ENVIRONMENTAL COMPLIANCE

The studies proposed provide for data collection and field sampling programs. No environmental effect of these programs occurs beyond that of traditional fisheries management data collection activities. These activities are within existing collecting permits or Federal special use permits issued to the Department of Fish and Game for scientific data collection. No other permits or other coordination activities are involved. This project received a categorical exclusion under the National

Environmental Policies Act.

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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, ADF&G
- 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.

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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
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, 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
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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
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C. Susan E. Merkouris, Fisheries Biologist II Commercial Fisheries Management and Development Alaska Department of Fish and Game Anchorage, Alaska 99518 (907) 267-2138

PROJECT RESPONSIBILITIES: Field coordination, sampling, archiving, contracting

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 Laboratory Technician, Microbiologist, Norton Sound Regional Hospital, Nome, AK
- SELECTED PUBLICATIONS AND PRESENTATIONS:
- Merkouris, S. E. and L. W. Seeb. (in prep). Biochemical genetic variation of highly exploited Tanner crabs, *Chionoecetes bairdi* and snow crabs, *C. opilio* in Alaska.
- 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.
- Bergstrom, D. J. et al. 1991. Annual Management Report Yukon Area. Alaska Department of Fish and Game, Regional Information Report Series.
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Date prepared

1996 EXXON VALDEZ TRU: COUNCIL PROJECT BUDGETOctober 1, 1*995 - September 30, 1996

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	Authorized	Proposed						
Budget Category:	FFY 1995	FFY 1996						
Personnel		\$20.8						
Travel		\$2.2						
Contractual		\$72.0						
Commodities		\$0.7						
Equipment		\$0.0		LONG F	RANGE FUNDI	NG REQUIREME	NTS	
Subtotal	\$0.0	\$95.7	Estimated	Estimated	Estimated	Estimated	Estimated	Estimated
General Administration		\$8.2	FFY 1997	FFY 1998	FFY 1999	FFY 2000	FFY 2001	FFY 2002
Project Total	\$0.0	\$103.9	\$120.0	\$97.0				
Full-time Equivalents (FTE)		0.3						
	I		Dollar amount	ts are shown in	thousands of	dollars.		
Other Resources							·	
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1996	Project Numb Project Title: Agency: AK	oer: 96165 Genetic dis Dept. of Fig	crimination c sh & Game	of PWS herrin	ng populatio	ns		FORM 3A AGENCY PROJECT DETAIL
Prepared: wh 2 Aug95 1 of	, L						j	8/2/95

1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET October 1, **1**995 - September 30, 1996

Pers	onnel Costs:		GS/Range/	Months	Monthly		Proposed
PM	Name	Position Description	Step	Budgeted	Costs	Overtime	FFY 1996
	Merkouris	FBII (PCN 1390)	16J	2.0	5,200		10.4
		FWT II	9 B	1.0	3,000		3.0
		,					0.0
*	Rosen	LIBI (PCN 6110)	17J	0.0	5,530		0.0
*	Hauser	FBIV (PCN 5103)	20M	1.0	7,432		7.4
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
l		Subtotal		4.0	21,162	0	
Tho	se costs associated with progr	am management should be indicated by place	ment of an *.		ł	Personnel Total	\$20.8
Trav	el Costs:		Ticket	Round	Total	Daily	Proposed
PM	Description		Price	Trips	Days	Per Diem	FFY 1996
l	Anchorage to Cordova-full fai	re	224	2			0.4
	Anchorage to Cordova-advan	ced purchase	70	4		1.5.0	0.3
	Per Diem				10	150	1.5
ļ			1				0.0
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Tho	se costs associated with progr	am management should be indicated by place	ement of an *.			I ravel I otal	\$2.2
r						۰ ۰	
		Breiset Numbers 06165					FORM 3B
	Personnel Personnel					Personnel	
	1990	Project Title: Genetic discrimination	n of PWS heri	ring population	ons		& Travel
Agency: AK Dept. of Fish & Game				DETAIL			

1996 EXXON VALDEZ TRUS COUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

Contractual Costs:		Proposed
Description		FFY 1996
Aircraft charter		1.5
Air Freight		0.5
DNA contract		70.0
When a non-truster	contractual Total	\$72.0
Commodities Costs	·	Proposed
Description		FFY 1996
Sampling supplies		0.7
	Commodities Total	\$0.7
· · · · · · · · · · · · · · · · · · ·		
	Project Number: 96165	
1006		ractual &
1990	Project litle: Genetic discrimination of PWS herring populations	modities
	Agency: AK Dept. of Fish & Game	FTÂII

1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

		Y		
New Equipment Pu	rchases:	Number	Unit	Proposed
Description		of Units	Price	FFY 1996
				0.0
	,			0.0
	``			0.0
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
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				0.0
				0.0
		1		0.0
Those purchases as	ssociated with replacement equipment should be indicated by placement of an R.	New E	quipment Total	\$0.0
Existing Equipment	Usage:		Number	Inventory
Description			of Units	Agency
				,
	¥ .			
	8			
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				,
	Project Number:			FORM 3B
1996	Project Title:		E	quipment
1000				DETAIL
	Agency: AK Dept. of Fish & Game			
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EXXON VALDEZ Oil Spill Trustee Council FY 96 Detailed Project Description

Herring Natal Habitats

Project Number:	96166
Restoration Category:	General Restoration
Proposer:	ADF&G
Lead Trustee Agency: Cooperating Agencies:	ADF&G University of Alaska
Duration:	Continue population monitoring until Pacific herring population in Prince William Sound has recovered; close out egg loss in FY96
Cost FY 96:	\$444,100
Cost FY 97:	\$405,600
Cost FY 98:	\$405,600
Cost FY 99:	\$280,000
Cost FY 00:	\$280,000
Cost FY 01:	\$280,000
Cost FY 02:	\$280,000
Geographic area:	Prince William Sound
Injured Resource/Service:	Pacific Herring

ABSTRACT

The 1989 Exxon Valdez oil spill coincided with the spring migration of Pacific herring *Clupea* pallasi 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 continue to provide estimates of spawning herring abundance through SCUBA and hydroacoustic studies, and to investigate the lethality of suspected pathogens and the role of environmental contaminants in disease transmission through laboratory and field studies.

INTRODUCTION

The Exxon Valdez oil spill coincided with the spring migration of Pacific herring *Clupea pallasi* 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. Oiling of over 40% of the spawning areas (42 of 98 miles used) caused elevated levels of physical and genetic abnormalities in newly hatched larvae and reduced hatching success of the embryos. 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.

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. Pathology studies from the spring of 1993 implicated viral hemorrhagic septicemia (VHS) as a potential source of mortality and stress. In 1994, the total observed spawning population was below threshold biomass required to conduct commercial harvest and no fishing occurred. Pathology studies indicated the presence of both VHS and a second potentially lethal pathogen, ichthyophonus. Pathology studies continued in 1995 and will include 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. The project will also develop embryo survival models that will improve the accuracy of spawn deposition biomass estimates. The project will be conducted in several parts. ADF&G will perform the field collection and data analysis constituting the continuation of herring spawn deposition surveys. A second field component will be continuating investigation of 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. The University of Alaska (UA) will perform 1) data analysis and modeling of egg loss data collected through 1995, 2) modeling of embryo survival, and 3) modeling of recruitment in relation to biological and environmental variables. A new component of this

project will be entry of aerial survey, harvest, spawn deposition, and egg loss data into a geographic information system (GIS) database.

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 by expanding echo integrated voltages by analytically determined target strengths. 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 examine issues relating to accuracy, reliability, and cost effectiveness of these methods.

A model which predicts egg loss will provide an indirect means to estimate the number of herring embryos physically removed from spawning areas by predation and wave action. Results from egg loss studies through 1995 will be used to identify important factors to include in models that can predict egg loss. Estimation of egg loss is useful for two purposes: (1) to improve the accuracy of spawn deposition biomass estimates by accounting for eggs lost between the time of spawning and the time of spawn surveys, and (2) to enable estimation of total embryo survival. Total embryo survival to the larval life stage will be used as the initial input of population abundance in life history models developed under project 96320, Sound Ecosystem Assessment (SEA). Data collected for this component of the current project will also be used to test hypotheses outlined in the Natal Habitat section of SEA.

Since it is not practical to measure all sources of egg morality each year, total embryo survival models will be used to relate mortality to more easily measured or estimated variables and characteristics of spawning habitat. Factors directly affecting the survival of embryos to larvae include wave action, predation, dessication during low tide, occurrence of cytogenetic abnormalities (which result in nonviable hatched larvae), pathogens, and pollution (which may elevate cytogenetic abnormality levels). These sources of direct mortality may be modified by environmental and biological variables such as wind direction, severity of storms, number of predators, availability of eggs to predators, type of substrate on which eggs are deposited, height of tidal fluctuation, water temperature, and air temperature. The degree to which these factors affect survival depends largely upon the characteristics of the habitat selected for egg deposition.

NEED FOR PROJECT

A. Statement of Problem

Adult Pacific herring on their way to PWS spawning area 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. 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. 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. 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.

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 would result 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 previously oiled areas. Additionally, there were approximately twice as many abnormal larvae from fish spawning in previously oiled areas.

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. In 1994, the total observed spawning population was below threshold biomass required to conduct commercial harvest and no fishing occurred. Pathology studies indicated the presence of both VHS and a second potentially lethal pathogen, ichthyophonus. Pathology studies have continued in 1995 and will include laboratory investigations of the lethality of suspect pathogens and the role of environmental contaminants in disease transmission.

B. Rationale

This project provides estimates of spawning herring abundance and a better understanding of some of the factors which contributed to the collapse of the population. This information is needed for monitoring recovery of the injured Prince William Sound (PWS) herring population. Project results can be used to judge recovery of the herring resource, including recovery to population levels sufficient for sustainable commercial harvest, and can also serve as the basis for setting harvest strategies. In addition, this project provides information about the abundance and survival of early life history stages which will improve our understanding of the ecological importance of herring in the PWS ecosystem.

C. Summary of Major Hypotheses and Objectives

The primary goal of this project is to monitor the recovery of Pacific herring which spawn within PWS, while a secondary goal is to improve our understanding of significant causes of mortality during early life history stages. Although project 96166 has not been included as a component of SEA, this secondary goal directly addresses the main hypothesis of the Natal Habitat portion of SEA which states that high energy coastal storms, temperature extremes, and predation control

density independent mortality and modify processes causing density dependent mortality of herring embryos. The effect of these physical and biological processes on the survival of the embryos varies with habitat.

The following subhypotheses are posed to direct the implementation of field work that will test components of this main conjecture:

- A. High energy storms cause formation of waves that physically remove eggs from herring spawning grounds. Waves remove eggs directly by dislodging them from vegetation to which they have been adhered and indirectly by dislodging vegetation containing attached eggs.
 - 1. Egg loss is positively correlated to the duration and intensity of wind-generated waves.
 - 2. Egg loss due to wave action is modified by the species of vegetation to which eggs are attached, the water depth in which eggs are deposited, and egg density.
 - 3. Site specific wave action is correlated with regional climatological conditions.
- B. Temperature extremes cause increased egg mortality. Elevated spring temperatures and increased ultraviolet radiation from increasing spring sunlight cause increased morphologic and cytogenetic abnormalities in herring embryos and reduce the number of viable larvae.
 - 1. Egg mortality in the intertidal zone increases with air temperatures $< 0 \circ C$ and $> 13.5 \circ C$.
 - 2. Egg mortality increases with continuous exposure to water temperatures $< 4 \circ C$.
 - 3. Incidence of cytogenetic and morphologic abnormalities and proportion of nonviable hatched larvae are increased at the upper and lower extremes of the ranges of temperature, salinity, and ultraviolet radiation typically occurring in PWS.
- C. Birds are the single most important predators on herring eggs.
 - 1. The distribution, timing, and abundance of gulls, seaducks, and shorebirds is positively correlated with the dispersion, timing, and abundance of herring spawn. Species composition of avian predators is dependent on spawn location and timing of spawn.
 - 2. Herring spawn is a major component in the diet of bird species foraging in herring spawn.
 - 3. Viable herring eggs are preferred prey compared to dead and decaying spawn.
 - 4. Avian consumption of spawn is greatest in the intertidal zone and varies with tidal height.
 - 5. Egg loss resulting from avian predation occurs at higher rates in years when eggs are scarce.

D. Completion Date

The egg loss component of the project will be completed in FY96. However, monitoring of the abundance, age composition and size composition of the PWS Pacific herring spawning population will be continued until the population has recovered.

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 pkrivate 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 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. Two vessels will be chartered as research platforms 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).

FY 96 BUDGET

Personnel	186.1
Travel	4.2
Contractual	199.5
Commodities	10.0
Equipment	2.4
Subtotal	402.2
Gen. Admin.	41.9
Total	444.1

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. The project has seven specific objectives in FY96:

- 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. Investigate the feasibility of estimating biomass of spawning herring using acoustic surveys and net sampling.
- 3. Compare estimates from spawn deposition with estimates from acoustic surveys.
- 4. Test a model of the relationship of spawn timing, spawner density and abundance to egg distribution and density.
- 5. Test a model relating sound-wide embryo survival to habitat utilized, egg density, and meteorological conditions.
- 6. Test a model relating historic recruitment success to biological and environmental variables.

While the overall goal of the project will not change in future years, specific objective might be altered to better achieve this goal. Estimation of the spawning herring population biomass, as well as its age, sex and size composition, will continue to be the most important objective of this project.

B. Methods

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 (Schwiegert et al. 1985) and Southeast Alaska (Blankenbeckler and Larson 1982, 1987). These surveys use random sampling for the first stage (transects) and systematic sampling for the second stage (quadrants within transects). Random sampling for the second stage is not feasible because of underwater logistical constraints (Schwiegert et al. 1985). Additionally, our surveys will be stratified by area to account for geographic differences and the potential of sampling discrete herring

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stocks. Areas surveyed may include Southeast, Northeast, North Shore, Naked Island and Montague Island (Figure 1).

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 quadrant, 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 quadrant placed at regular intervals along the length of the transect. The sampling quadrant will consist of a 0.1 m² PVC pipe frame with a depth gauge and compass attached. The first quadrant location will be randomly selected within the first 5 meters of spawn. Succeeding quadrant locations will be systematically spaced every 5 meters along the compass course until the apparent end of the spawn is found. Within each quadrant, the lead diver will estimate the number of eggs in units of thousands (K) within the quadrant, 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 quadrant. All egg-containing vegetation within the quadrant 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 80 calibration samples will be needed for each uncalibrated diver (less than three years survey participation) and 40 for each calibrated diver (three or more years survey participation). One quarter of the total samples will be taken for each of the four vegetation categories: eelgrass (EEL), fucus (FUC), large brown kelp (LBK), and hair kelp (HRK). One third of the calibration samples will be stratified over three ranges of egg densities: low (0-20,000), medium (20,000-80,000), and high (> 80,000) within each vegetation category. 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), 1989-1992 (Biggs et al. in press). The biomass estimator will be

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

where

B	=	estimated spawning biomass in tonnes,
Т	=	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)

$$Var(B) = T^{2}Var(B') + B^{2}Var(T) - Var(T)Var(B'),$$
⁽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 Schwiegert 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 m² 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

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

where

L	=	the shoreline length of the spawn-containing stratum in meters,
Ν	=	$L/0.1^{0.5}$ = the total number of possible transects,
0.1 ^{0.5}	=	0.3162 m = width of transect strip,
ŷ	=	average estimated total number of eggs (thousands) per transect,
10-6	=	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

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

where and -

n	=	number of transects actually sampled,
i	=	transect number,
Mi	=	$w_i/0.1^{0.5}$ = number of possible quadrants in transect i,
w _i		spawn patch width in meters measured as the distance along the transect between the
		first quadrant containing eggs and the last quadrant containing eggs, and
$\overline{\mathbf{y}}_{i}$	=	average quadrant egg count in transect i (in thousands of eggs).

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

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

where

j	Ħ	quadrant number within transect i,
m _i	=	number of quadrants actually sampled in transect i, and
y _{ij}	=	adjusted diver-estimated egg count (in thousands of eggs) from the diver calibration
-		model for quadrant 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 (quadrants), 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

$$[N^{2}(10^{-6})^{2}[\frac{(1-f_{1})}{n}s_{1}^{2} + \frac{f_{1}(1-f_{2})}{n}s_{2}^{2} + \frac{f_{1}f_{2}}{n}s_{3}^{2}]]$$

$$Var(T) = \frac{\sum_{i=1}^{n} m_{i}}{(1-R)^{2}},$$
(7)

where

variance among transects,

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

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

variance among quadrants,

$$s_{3}^{2} = \sum_{i=1}^{n} \sum_{j=1}^{m_{i}} Var(y_{ij}) =$$
(10)

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

$$f_1 = \frac{n}{N} = \tag{11}$$

proportion of possible transects sampled, and

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

proportion of quadrants 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 quadrant 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 1994 calibrations (personal communications. Ed Debevec, ADF&G, Cordova). 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

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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 will be 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

$$B' = \frac{\overline{WS}}{F(\overline{W}_{f})} 10^{3}, \tag{13}$$

where

- \vec{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,
- $F(W_i)$ = estimated fecundity at the average weight of females in the spawning population in an area, in numbers of eggs, and

$$\frac{10^3 = \text{ conversion factor}}{10^9} = \frac{10^{-6}}{\text{eggs to billions}}$$

Because average weight, sex ratio and fecundity will be all estimated from the same herring samples, the estimates will be not independent. The variance of B' is approximately:

$$Var(B') = (10^{3})^{2} \left[\left[\frac{S}{F(\overline{W}_{f})} \right]^{2} Var(\overline{W}) + \left[\frac{\overline{W}}{F(\overline{W}_{f})} \right]^{2} Var(S) + \left[\frac{\overline{W}S}{F(\overline{W}_{f})^{2}} \right]^{2} Var(F(\overline{W}_{f})) + 2Cov(\overline{W}, S) \left[\frac{S}{F(\overline{W}_{f})} \right] \left[\frac{\overline{W}}{F(\overline{W}_{f})} \right] \left[\frac{\overline{W}}{F(\overline{W}_{f})} \right] \left[\frac{S}{F(\overline{W}_{f})} \right] \left[\frac{\overline{W}S}{F(F(\overline{W}_{f}))^{2}} \right] - 2Cov[\overline{W}, F(\overline{W}_{f})] \left[\frac{\overline{W}}{F(\overline{W}_{f})} \right] \left[\frac{\overline{W}S}{F(\overline{W}_{f})} \right] \left[\frac{\overline{W}S}{F(\overline{W}_{f})} \right] \left[\frac{\overline{W}S}{F(\overline{W}_{f})^{2}} \right]$$

$$(14)$$

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, $Cov(\overline{W},S)$ and $Cov[S,F(\overline{W}_f)]$. Because the term involving $Cov[\overline{W},F(\overline{W}_f)]$ 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, 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. Because commercial herring fishing not likely to be open again in 1996, AWL samples will 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 ($F(W_f)$) from expression (12).

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. 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

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

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

Fecundity for Biomass Estimates. Average fecundity for PWS will be estimated from a fecundityweight relationship as $F(\overline{W_t})$, and used in equation 12 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)

$$Var[F(\bar{W}_{f})] = s^{2} \left[\frac{1}{n} + \frac{1}{q} + \frac{(\bar{W}_{f} - \bar{W}\bar{F})^{2}}{\sum (W_{i} - \bar{W}\bar{F})^{2}}\right],$$
(16)

where

s	=	the residual mean square from the fecundity-weight linear regression,
$\bar{W_f}$	=	the average weight of female fish in the spawning population,
WF	=	the average weight of females in the fecundity sample,
Wi		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 relationships 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 96320 (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.

Acoustic Survey and Biomass Estimation

Standard acoustic techniques (Urick 1975; Thorne 1983b; Ehrenberg and Lytle 1972) for echointegration and dual beam processing of target strength will be used to independently estimate

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the biomass of herring present near spawning grounds during the spring migration. Energy reflected from fish concentrations will be measured and converted to fish density using measurements of energy reflected from single fish (target strength) and knowledge of the sample volume (transducer directivity). Net sampling will be conducted to subsample the acoustic targets to verify species, size and obtain other biological information on the insonified fish (Thomas 1992).

The acoustic survey will employ one commercial purse seiner under short term vessel charter to assist in searching for herring schools and to conduct net sampling. The scientific echosounding equipment will be located aboard the ADF&G research vessel *Montague* for acoustic mapping of biomass. The acoustics vessel will be outfitted with either a BioSonics 70 or 120 Khz echo sounder with a dual beam pre-amplified transducer mounted on a 1.2 m BioSonics Biofin in a down-looking configuration. The Biofin will be towed at a depth of about 2 m at approximately 5 m off to one side of the vessel. The catching vessel will be equipped with a seine approximately 30 m deep typical of the gear-type used in the commercial sac roe herring fishery.

Survey Design. The acoustic survey will be a multistage sampling design (Cochran 1967). Historical information about location of spawning, aerial surveys of herring schools, and wide scale searches using ship's searchlight (sweeping) and down-looking echosounders will be used to locate concentrations of herring schools in a first stage search. The second stage of sampling will be to map school groups and measure the density using the scientific echosounder. Acoustic survey transects will be run in a zigzag fashion over the school groups and will be replicated during both day and night for large school groups.

Acoustic Parameters. Target strength information for herring will be derived from average length to target strength (in decibels) per kg fish after Thorne (1983a). Thorne's (1983a) empirical relationship assumes the following logistical equation:

$$\gamma = \frac{\overline{\sigma}}{\overline{W}} = a_{\bullet}^{\overline{\tau} \cdot h}$$
(17)

where σ is the mean acoustic backscattering coefficient, W is the mean weight (in kg), 1 is the mean length (in cm), and a and b are constants. Values for the constants (a and b) are obtained from data for a variety of fisheries presented by Thorne using a linear regression of \log_{10} l versus 10 log (σ /w), where 10 log (σ /w) is referred to in Thorne (1983a) as "target strength per kg." Average herring length and weight data will be compiled from samples obtained by the purse seine catcher vessel. These measured data will be applied to Thorne's (1983a) empirical relationship to obtain the ratio γ = σ /w and the mean backscatter coefficient (σ). As a cross check, *in situ* measurements of target strength from dual beam acoustic data will be generated and compared with Thorne's (1983a) empirical formula.

Biomass estimation. Herring biomass will be calculated for each zigzag survey. The general calculation of the population density using echointegration for a single cell jk on a transect is given as

$$\beta_{jk} = \rho_{jk} \,\overline{w}_{jk} = \frac{C(ei)_{jk} \cdot P_{jk}}{\frac{\overline{\sigma}_{jk}}{\overline{w}_{jk}}}$$
(18)

where β_{jk} is the population density (mass per unit volume), ρ_{jk} is the density of scatterers, w_{jk} is mean weight of scatterers, C is acoustic constant (calibration settings ie., gain etc.) e_{ijk} is the mean of the voltage squared, P_{jk} is percentage of cell *jk* within the water column, and σ_{jk} is mean backscattering coefficient for targets within cell *jk*.

The biomass for a region of surface area A is determined by using a set of line transects along which a total of nrs point estimates of biomass per unit area is obtained. Specifically,

$$B = \frac{\sum_{j=1}^{nrs} \sum_{k=1}^{nst} \beta_{jk}}{nrs} \cdot A$$
(19)

where nrs is number of reports (along the line transects), nst is number of depth strata, and A is survey area.

Herring biomass estimates will follow Thorne (1983a), assuming that σ_{jk}/w_{jk} is independent of cell *jk*, hence, for all *jk* σ_{jk}/w_{jk} is a constant γ , and γ is given by equation 1. With this assumption, equation 4 simplifies to:

$$\beta_{jk} = \frac{C}{\gamma} (ei)_{jk} P_{jk}$$
(20)

and the herring biomass B in an area is given as

$$B = \frac{C}{\gamma} \frac{\sum_{j} \sum_{k} (ei)_{jk} P_{jk}}{nrs} A$$
(21)

Egg Loss Study

Analysis of previously collected information on the proportion of eggs lost through physical removal and the mortality rate of remaining eggs will be summarized and reported in FY96. Important factors in egg loss will be identified through this analysis and used to guide future study design to estimate egg loss indirectly through modeling and annual measurement of these important factors.

Prior to 1994, an assumed constant of 10% egg loss for surveys generally conducted 5-6 days after spawning was used based upon values recommended in the literature (Haegele et al. 1981, Blankenbeckler and Larson 1982). Other investigators (J. Schweigert, personal communication,

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Canadian Department of Fisheries and Oceans, Nanaimo, B.C.; Biggs et al. *in press*) estimated egg loss rates, but did not include collection of data to relate egg loss to habitat, environmental conditions or predation. Beginning in 1994, egg loss study design for this project has included modifications to identify important factors in egg loss to facilitate modeling. Identification of important factors included measurements to describe habitat characteristics, physical and oceanographic environmental conditions, and avian predation. In 1995, wave energy at spawning sites and local meteorological conditions were also recorded in conjunction with egg loss estimates.

Measurements of physical conditions and observations of habitat characteristics gathered for egg loss sites during previous field seasons will be tested for correlations with meteorological conditions and with rates of egg loss to address hypotheses A and B. Physical measurements and observations included air and water temperature, salinity, precipitation, wind speed and direction, wave height and wave direction and were collected by survey divers during each egg loss site visit. Gradient, substrate and vegetation were collected at each site during setup. Electronic recording instruments were also used to gather data on environmental conditions including temperature, wave height, and tide height at fixed locations during incubation. The University of Alaska will incorporate these physical and meteorological data into egg loss and embryo survival models.

For the current proposed project, regional meteorological and oceanographic data will be gathered from shipboard surveys, moored instrumentation, and existing data products from government agencies. Project personnel will work with SEA investigators for the acquisition and archival of data products. These measurements will be used to model the effect of meteorological conditions on wave activity and the resulting effects on egg loss and embryo survival. Data analysis will be the primary responsibility of the University of Alaska with assistance and support from project personnel and SEA investigators.

Egg Loss Data Analysis. Development and selection of appropriate statistical analyses for egg loss are currently in progress. If no refinements to previous techniques are deemed appropriate, an exponential decay model will be used to estimate loss in numbers of eggs over time for bias corrected similar to that used for the 1990 and 1991 data:

$$ADJ_{ijk} = e^{\alpha} e^{trans_j} e^{depth_k} e^{\tau_{jk}(days_{ijk})} e^{\epsilon_{ijk}}, \qquad (22)$$

where

α	=	a constant,
ADJ _{ijk}	=	adjusted egg density estimates,
transi	=	parameters representing the effect of transect j,
depth _k	=	parameters representing the effect of depth k,
$ au_{jk}$		parameters controlling the functional form of the relationship between egg density and time (number of days after spawning),
days _{ijk}	-	the number of days after spawning occurred, and
ε _{ijk}	=	normally distributed random variable with mean = 0 and variance = σ^2 .

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A multiplicative model will be chosen because egg numbers will be expected to vary with location (transect) and depth. All interactive terms will be included in the model. After a logarithmic transformation, equation 22 became

$$\log_{e}(ADJ_{ijk}) = \alpha + trans_{j} + depth_{k} + \tau_{jk}(days_{ijk}) + \epsilon_{ijk}.$$
(23)

In logarithmic form, the model comprised a linear analysis of covariance (ANCOVA) with two factor effects (transect and depth) and 1 covariate (number of days after spawning). SAS (1987) procedure for general linear models (GLM) will be used to obtain least squares estimates of the parameters. Estimates of eggs over time (days) were then made for each transect and depth.

The egg survival model used to track the data collected in 1989 through 1991 in PWS took the form of the following analysis of covariance (ANCOVA)

$$\begin{aligned} \arcsin(s) &= \mu + treat_{j} + depth_{k} + day_{i} + treat * depth_{jk} + \\ day * treat_{ij} + day * depth_{ik} + day * treat * depth_{ijk} + \\ trans(treat)_{l(i)} + \epsilon_{ijkl}. \end{aligned}$$
(24)

Future analyses may include replacing the treatment term used to differentiate between oil and control areas with a treatment term for habitat type. The egg loss and current egg survival models will eventually be synthesized into an embryo survival model that incorporates habitat type, predation, and the relationship of meteorological conditions on wave action. The ultimate goal, as outlined in the NHP portion of the SEA plan, will be to build a sound-wide embryo survival model relating habitat type, egg density, predation, and meteorological conditions.

Systematic bias in diver estimates at egg loss sites will be assumed to be the same as diver estimates for spawn deposition surveys and the model used will be identical.

C. Contracts and Other Agency Assistance

Through a competitive bidding process, one or more purse seine vessels will be chartered to capture fish for species and size composition of acoustic targets, AWL/fecundity samples, spawning adult herring for histopathology samples (project 96320S), and reproductive impairment samples for (project 96074). Depending upon the duration of the work and other competing uses, the ADF&G R/V Montague may be used as a sampling platform and as a scientific acoustics vessel either at no charge or at a standard rate of \$1,200/d. 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 mid-April.

Two vessels will be chartered through a standard competitive bid process as research platforms for spawn deposition surveys. These vessels 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.

Biometric and computer programming assistance for egg loss and recruitment studies will be contracted with the University of Alaska through a Reimbursable Services Contract.

D. 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. Another benefit of the project will be the information gained during egg loss studies which may be extremely valuable in assessing critical habitat and energy needs of migratory birds using PWS.

SCHEDULE

A. Measurable Project Tasks for FY 96

December	Finalize FY96 Detailed Project Description
February	1995 Biomass estimates - Dept. Forecast and Stock Assessment Reports
April	Before onset of spawning:
-	Conduct acoustic survey (5-7 d)
	Collect AWL, fecundity, disease, genetic stock ID, and bioenergetics samples
	After onset of spawning:
	Initiate dive surveys
	Assist reproductive impairment sample collection
May	Submit FY97 Draft Detailed Project Description
•	Complete dive surveys
	Begin lab processing of diver calibration calibration and fecundity samples
June	Complete calibration sample processing samples
September	Finalize estimate of spawning biomass
November	Finalize projection of 1997 run biomass
April 1997	Submit FY96 annual report - biomass estimate
June 1997	Submit Final report for Egg Loss study

B. Project Milestones and Endpoints

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

September 1996	Objective 1:	Finalize estimate of spawning biomass of herring in 1996 using spawn deposition methodology.
October 1996	Objective 2:	Finalize acoustic estimate of spawning biomass of herring in 1996 & decide whether to continue or modify program.
December 1996	Objective 3:	Test egg distribution\density model
January 1997	Objective 4:	Test embryo survival model
February 1997	Objective 5:	Test recruitment success model

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C. Project 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 Board of Trustees and Chief Scientist. Significant findings presented in status reports and final reports will be submitted for publication in peer reviewed journals and presentation at scientific symposia as they are obtained. A final report will be submitted for FY96 by April 15, 1997.

COORDINATION AND INTEGRATION OF RESTORATION EFFORT

Project 96166 will be integrated closely with project 96320, 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 (96320T) will require location and abundance of spawn as well as information about age and size structure of sampled catches. Physical measurements taken for project 96166 may be useful to project 96320M. Information about spawn distribution will also be useful in drafting a study design for herring larval advection studies.

Project 96166 will also share information and resources with Project 96165, Herring Genetic Stock Identification in PWS. Additional samples required for this project beyond FY96 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 96166 for sample collection include Reproductive Impairment (96074), Somatic and Spawning Energetics of Herring/Pollock (96320U), and Disease Impacts on PWS Herring Populations (96320S).

Finally, integration of research will require data sharing and coordination with Project 96163, Forage Fish Influence on Injured Species. Herring are an important forage 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.

ENVIRONMENTAL COMPLIANCE

These activities are within existing collecting permits or Federal special use permits issued to ADF&G for scientific data collection. This project received a categorical exclusion under the National Environmental Policies Act (NEPA). Federal OSHA regulations covering hazardous materials handling and disposal, and lab safety training for personnel working with preservation chemicals will be followed. No other permits or other coordination activities are involved.

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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: 1985Master 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 sockeve 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. 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, 1995); R.T. Cooney, T.M. Willette, and S. Sharr. 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. (1992). Willette, T.M. and R.T. Cooney. 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, (1991). Eggers, D.M., L.R. Peltz, B.G. Bue, and T.M. Willette. 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. (1991). Member: American Fisheries Society, Alaska Chapter.

Evelyn D. (Biggs) Brown (Co-investigator), M.S., Herring Fisheries Research Biologist, Alaska Department of Fish and Game, P.O. Box 669, Cordova, Alaska 99574. Education: Masters of Science, Fisheries and Aquacultural Engineering, Oregon State University, 1980: Bachelors of Science, Zoology and Chemistry, University of Utah, 1977. Professional Experience: Herring Research Project Leader, ADFG, 1988-1993; Sonar Project Leader-Mullet Project, Florida Department of Natural Resources, 1987-1988; Sonar Project Leader-Copper River, ADFG, 1985-1987; Marine Biologist-Shipboard Duty, NOAA, 1983; Fisheries and Marine Biologist for Metlakatla Indian Community, Annette Island, Alaska, 1980-1982. Research Projects: Principal Investigator for Injury to PWS Herring After the Oil Spill, 1989-1993; Spawn Deposition Survey-Underwater Research Program, 1988-1992; Mullet Study using Hydroacoustics, Manistee River, Florida, 1987-1988; Miles Lake Salmon Enumeration Sonar, 1985-1987; Marine Mammal-Japanese Fleet Interaction Research, 1983; Annette Island Crab and Abalone Subsistence Harvest Plan, 1981; Annette Island Environmental Impact Statement for Timber Harvest Activities, 1981-1982: Annette Island Herring Management Plan, 1981-1982; Annette Island Salmon Stream Inventory and Recommended Escapement, 1981-1982; Annette Island Oyster Culture Commercial Feasibility Project, 1980-1981; Selected Publications: Biggs, E.D. et al. The Exxon Valdez oil spill and Pacific herring in Prince William Sound: a summary of lethal. sublethal and long-term effects from 1989-1993. In: Proceedings of the Exxon Valdez Oil Spill Symposium, American Fisheries Society Symposium Series, (in press, 1993); Biggs, E.D. and F. Funk, Pacific herring spawning ground surve for Prince William Sound, 1988, with historic overview. ADFG Regional Informational Repo., 2C88-07. Anchorage, Alaska. 45 p (1988). Member: American Fisheries Society, Alaska Chapter.

Ed Debevec (Project Biometrician), CFMDD, ADF&G, P.O. Box 669, Cordova, Alaska 99574. Education: Master of Science, Statistics, Oregon State University, 1994; Bachelor of Science, Biological Sciences, University of Alaska Fairbanks, 1991. Professional Experience: Alaska Department of Fish and Game since November 1994. Data analysis and study design for Prince William Sound pink salmon predator study. Selected Publications: Debevec, E.M. and S.F. MacLean, Design of greenhouses for the manipulation of temperature in tundra plant communities, Arctic and Alpine Research (1993). Armbruster, W.S., M.E. Edwards, and E.M. Debevec, Floral character displacement generates assemblage structure of western Australian triggerplants (Stylidium), Ecology (1994). Mark Willette, Principal Investigator Area Research Biologist Comm. Fisheries Management and Development Div. (CFMDD) ADF&G, Cordova, Alaska

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Fisheries Program Manager Habitat and Restoration Div. ADF&G, Anchorage, Alaska

Date Prepared

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

budget Category:	Authorized FFY 1995	Proposed FFY 1996						
Personnel	\$195.7	\$0.0						
Contractual	\$4.0	<u> </u>						
Commodition	\$248.0	\$2.0						
Equipment	¢11.3	0.0					NITC	
Subtatal	\$0.1 \$466.1	40.0 62.8	Entimated	Ectimated	Estimated	Estimated	Ectimated	Estimated
General Administration	\$400.1	\$2.0	ESUMATED	ESUITATEU	ESUMALOU	ESTIMATED	ESUMALEU EEV 2001	FEX 2002
Broiget Total	\$40.7	÷7.2	6405 G	¢405.6	6290 O	\$280.0	\$280.0	\$280.0
Project rotal	\$512.0	92.0	\$405.0	\$405.0	\$280.0	\$200.0	\$200.0	\$280.0
Full-time Equivalents (FTE)	3.2	0.0						
			Dollar amount	s are shown in	thousands of c	ollars.		
Other Resources							3	
After FY98, a decision will be	made to continue ei	ther spawn de	position or hyd	roacoustic biom	nass assessmen	ts at a reduced	cost.	
After FY98, a decision will be	made to continue ei	ther spawn de	position or hyd	roacoustic bion	nass assessmen	ts at a reduced	cost.	
After FY98, a decision will be	made to continue ei	ther spawn de	position or hyd	roacoustic bion	nass assessmen	ts at a reduced	cost.	

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

Pers	onnel Costs:		GS/Range/	Months	Monthly	1	Proposed
PM	Name	Position Description	Step	Budgeted	Costs	Overtime	FFY 1996
*							0.0
*				Į			0.0
		`		*			0.0
	•						0.0
							0.0
		۲ ۰					0.0
-							0.0
		1 ⁹					0.0
							0.0
							0.0
							0.0
							0.0
						۶.	0.0
	L	Subtotal		0.0	0	0	
Tho	se costs associated with progr	am management should be indicated by place	ment of an *.		F	Personnel Total	\$0.0
Trav	el Costs:		Ticket	Round	Total	Daily	Proposed
PM			Price	Trips	Days	Per Diem	FFY 1996
l	Description						0.0
							0.0
							0.0
							0.0
							0.0
l							0.0
1							0.0
							0.0
							0.0
Tho	se costs associated with progr	am management should be indicated by place	ment of an *.			Travel Total	\$0.0
l ine and							
	T						FORM 3B
		Project Number: 96166 (Interim)					Personnel
	1996	Project Title: Herring Natal Habitats					& Traval
		Agency: AK Dept. of Fish & Game					
							DETAIL

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

Contractual Costs:			Proposed
Description			FFY 1996
Dive master class (CPR training (4 clas Dive physicals	(1 class @ \$400) sses @ \$100 per class)	*	0.4 0.4 1.8
When a non-trustee orga	anization is used, the form 4A is required.	Contractual Total	\$2.6
Commodities Costs:			Proposed
Description			FFY 1996
	:		
		Commodities Total	\$0.0
			and a contract "Inter Painty, and a
=	Project Number: 96166 (Interim)		ORM 3B
1996	Project Title: Herring Natal Habitata	Co	ntractual &
	Ageneyy AK Dept of Fick 9 Come		ommodities
	Agency: AK Dept. of Fish & Game		DETAIL

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

New	/ Equipment Purchases:	Number	Unit	Proposed
Desc	cription	of Units	Price	FFY 1996
				0.0
				0.0
				0.0
				0.0
	i			0.0
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
Thos	se purchases associated with replacement equipment should be indicated by placement of an R.	New E	quipment Total	\$0.0
Exis	ting Equipment Usage: (partial inventory)		Number	Inventory
Desc	cription		of Units	Agency
	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)			
			<u>г</u>	
	Project Number: 96166 (Interim)			FORM 3B
	1996 Project Title: Herring Natal Habitats		E	quipment
1	Agenery, AK Dent, of Eich & Geme			DETAIL
	Agency: AN Dept. of Fish & Game		Ì	
L				7/04/05

4 of 4

7/24/95

1996 EXXON VALDEZ TRUL. __ COUNCIL PROJECT BUDGET October 1, 1995 - September 30, 1996

4

Budget Category:	Authorizeu	Proposed						
	FFY 1995	FFY 1996						
	· · ·							
Personnel	\$195.7	\$83.5						
Travel	\$4.8	\$2.7						
Contractual	\$248.6	\$119.3						
Commodities	\$11.9	\$0.7						
Equipment	\$5.1	\$0.0		LÓNG F	RANGE FUNDIN	g requiremen	NTS	
Subtotal	\$466.1	\$206.2	Estimated	Estimated	Estimated	Estimated	Estimated	Estimated
General Administration	\$46.7	\$20.9	FFY 1997	FFY 1998	FFY 1999	FFY 2000	FFY 2001	FFY 2002
Project Total	\$512.8	\$227.1	\$405.6	\$405.6	\$280.0	\$280.0	\$280.0	\$280.0
Full-time Equivalents (FTE	3.2	1.3						
			Dollar amount	s are shown in	thousands of d	ollars.		
Other Resources							`	
After FY98, a decision w	III be made to continue (either spawn de	position or hyd	roacoustic bion	nass assessmen	ts at a reduced	i cost.	
After FY98, a decision w		either spawn de	position or hyd	roacoustic bion	nass assessmen	ts at a reduced	i cost.	
After FY98, a decision w		either spawn de	position or hyd	roacoustic bion	nass assessmen	ts at a reduced	i cost.	
After FY98, a decision w		either spawn de	position or hyd	roacoustic bion	nass assessmen	ts at a reduced	l cost.	41

1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

Per	sonnel Costs:	GS/Range/	Months	Monthly	T	Proposed	
PM	Name	Position Description	Step	Budgeted	Costs	Overtime	FFY 1996
*	Vacant- Hauser	Program Manager	20L	1.0	7,182	0	7.2
	M. Willette	Fishery Biologist III	18D	3.0	5,866	0	17.6
	E. Brown	Fishery Biologist IL	16E	¥ 6.0	5,282	0	31.7
	E. Debevec	Biometrician I	17A	5.0	4,753	0	23.8
	Vacant	Field Office Assistant	9A	1.0	3,200	0	3.2
		Subto	tal	16.0	26,283	0	
The	se costs associated with	program management should be indicated by pla	acement of an *.		Pe	rsonnel Total	\$83.5
Tra	vel Costs:		Ticket	Round	Total	Daily	Proposed
PM			Price	Trips	Days	Per Diem	FFY 1996
	Description	i i i i i i i i i i i i i i i i i i i	and the second s				0.0
		•					0.0
	RT Cordova-Anch., Atte	and herring synthesis meeting, 2 staff	200	2	4	95	0.8
	RT Cordova-Anch., Atte	and EVOS annual workshop, 2 staff	200	2	7	95	+ 1.1
	RT Cordova-Anch., Atte	and meeting with herring mgmt staff, 2 staff	200	2	4	95	0.8
							0.0
		· · · · · · · · · · · · · · · · · · ·					0.0
			a start and a start				0.0
			The strend of	2 2 2			0.0
The		are green management should be indicated by pl	nonmant of an t	and the second		Travel Total	62.7
Inc	se costs associated with	program management should be indicated by pla	acement of all .			Traver Total	92.7
	T					E	ORM 3B
	a strategy of	Project Number: 96166 (Report V	Vriting)			D	orini ob
	1996	Project Title: Herring Natal Habita	ts				ersonner
		Agency: AK Dent of Fish & Gam	0			8	k Iravel
		Agency. An Dept. of Fish & Gam	6				DETAIL

1996 EXXON VALDEZ TR:E COUNCIL PROJECOctober 1, 1995 - September 30, 1996 E COUNCIL PROJECT BUDGET

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Contractual Costs:			Proposed
Description			FFY 1996
Publication costs			2.3
Network operation & n	naintenance		0.5
Contract for GIS spaw	n mapping	a'	36.0
Contract for egg loss a	nd recruitment model (UAF)		80.0
Hazmat disposal			0.5
When a non-trustee organiz	ation is used, the form 4A is required.	Contractual Tota	\$119.3
Commodities Costs:			Proposed
Description	· · · · · · · · · · · · · · · · · · ·	· · · · · · · · · · · · · · · · · · ·	FFY 1996
Software upgrades Office supplies			0.2
		Commodities Total	\$0.7
[]F			FORM 3B
	Project Number: 96166 (Report Writing)		ntractual &
1996	Project Title: Herring Natal Habitats		
	Agency: AK Dept. of Fish & Game		
	5		DETAIL
3 (of <i>f</i> 4		7/24/95

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

New	Equipment Purchases:		Number	Unit	Proposed
Desc	cription		of Units	Price	FFY 1996
					0.0
					0.0
		۲. ۲			0.0
	•				0.0
					0.0
					0.0
					0.0
					0.0
					0.0
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					0.0
	l		1		0.0
Tho	se purchases associated with r	eplacement equipment should be indicated by placement of an R.	New E	quipment Total	\$0.0
Exis	ting Equipment Usage: (partia	l inventory)		Number	inventory
Des	cription			of Units	Agency
	Manual and Alde second as with			2	
	Alumnused Can Dany (20 ft)	computer interface		3	ADro
1	Alumaweid Sea Dory (20 II)			1	
	Boston Whater (17 ft)			20	
	Dive regulators			29	
	Dive depth/pressure gauges	,		14	
	Dive backpacks				
	Dry guite (various sizes)			14	r
	Dive tanks			21	
	Dive Laiks			~ 1	
	(complete inventory on file at	ADEG Cordova)			
1	(complete inventory on me at				
			<u></u>		
	1000	Project Number: 96166 (Report Writing)			
1	1996	Project Title: Herring Natal Habitats			quipment
		Agency: AK Dept. of Fish & Game		`	DETAIL
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Isotope Ratio Studies of Marine Mammals in Prince William Sound

Project Number:	96170
Restoration Category:	Research
Proposer:	University of Alaska Fairbanks
Lead Trustee Agency:	Alaska Department of Fish and Game
Cooperating Agencies:	-
Duration:	3 Years
Cost of FY 96:	\$128,400
Cost of FY 97:	\$130,000
Cost of FY 98:	\$110,000
Cost of FY 99:	
Cost of FY 00:	
Cost of FY 01:	
Cost of FY 02:	
Geographic Area:	Prince William Sound/Gulf of Alaska
Injured Resource/	
Service:	Harbor seals, nearshore ecosystem species, seabirds

ABSTRACT

This project contributes to the studies by the ADF&G personnel to determine the reasons for the decline of harbor seal and steller sea lion populations in Prince William Sound (PWS). In addition, it contributes to the SEA program being conducted by the Prince William Sound Science Center and the Institute of Marine Science, UAF, to describe the food chains supporting important commercial fish species impacted by the Exxon Valdez Oil Spill. We seek to better describe the trophic interactions and trophic status of marine mammals, birds and their prey species. The integrating methodology for this wide range of tasks is the use of stable isotope ratios as natural tracers of carbon and nitrogen transfers through the food webs. Through a mix of captive animal studies, comparison of isotope ratios in archived and current marine mammal tissues and their potential prey species in the PWS, insight into environmental changes causing the decline may be possible. We also will supply the isotope ratio determinations for other projects using this technique in the PWS ecosystem. Over the 12 months of FY96 funding we anticipate the analysis of approximately 10,000 samples in these related projects.

INTRODUCTION

Over the past two decades, isotope ratio analysis has emerged as a powerful tool in ecosystem research both on the process scale and as a validation technique for large-scale ecosystem models (Michener and Schell, 1994). In relevant applications to this study, Schell et al (1989a) described a geographic gradient in isotope ratios in biota across the Alaskan Beaufort Sea and the Bering-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 clear up to the top predators. Saupe et al. (1989) describes the parallel shifts in δ^{13} C in euphausiids and copepods across this region and Schell et al (1989b, 1993) 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 $\delta^{15}N$ of euphausiid and copepods in the Bering, Chukchi and Beaufort seas are statistically indistinguishable but that when pelagic and benthic species of known feeding habits are compared, a predictable enrichment in ¹⁵N occurs of about 3.3% per trophic level increase. By assembling the trophic spectrum of species within an ecosystem it is possible to ascribe trophic status within the ecosystem. Hobson and Welch (1992) were able to used $\delta^{15}N$ values in the Barrow Strait - Lancaster Sound region to identify the roles of arctic cod (Boreogadus saida) and other prey species to top consumers. Higher trophic levels showed little change in δ^{13} 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. Schell (in press) has further shown that in the Bering sea lions are not as high in trophic status as spotted seals and span a wide trophic range between individuals. Whereas an individual animal may show little temporal change in trophic status as indicated by $\delta^{15}N$ 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 few individuals examined to date. In comparing vibrissae with several years of growth, it is readily evident that some sea lions feed over at full trophic levels apart. This study expands upon our growing data base to provide a similar assessment of the trophic energetics of harbor seals in Prince William Sound and to assist other EVOS studies by providing isotope ratio analysis and interpretation for their studies.

Funding for this work resumed in February 1995 (no interim funds from Oct. 94 were allocated) and as a result only preliminary findings are available at this time. 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 the Alaska Department of Fish and Game. We have met with the chief veterinarian of the Mystic Marine Life Aquarium and they have kindly agreed upon protocols for undertaking experiments on captive sea lions and harbor seals aimed at determination of whisker growth rates, diet fractionation factors arising from differing types of prey species, and seasonal cycles in isotope ratios arising from physiological effects. 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.

NEED FOR THE PROJECT

A. Statement of the 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 relationships have 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 past decade as underlying reasons for the decline.

A second need for this project is to provide isotope ratio analyses for this study and other restoration projects needing isotope abundance information. We anticipate receiving a total of approximately 10,000 samples for isotope ratio analysis in the coming year.

B. Rationale

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.

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 ä with accompanying loss of the lighter isotope through excretion. The enrichment occurs with each trophic step and thus 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 thus 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, macrozooplankton 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. Summary of Major Hypotheses and Objectives

The major hypothesis to be tested is that:

The isotope ratios of harbor seals are derived from prey taken from the Prince William Sound ecosystem. Changes in the trophic structure of the food webs arising from either the oil spill or natural cycles will be evident through changes in isotope ratios in potential prey and seals.

The objectives of this study are divided into three elements:

- 1. A research component on marine mammals focusing on the trophic energetics and ecosystem dynamics of harbor seals conducted by Dr. Schell, PI, in cooperation with ADF&G personnel working as part of the marine mammal program. An additional effort using captive animals to calibrate the response to changing isotopic composition in diet and to determine vibrissae growth rates will also be undertaken. This will entail the analysis of approximately 3000-4000 samples for carbon and nitrogen isotope ratios, a major increase over last year reflecting the very successful collection of samples from over 100 seals.
- 2. A research component focusing on lower trophic levels having direct application to the testing of hypotheses regarding fisheries resources. This work will be conducted in cooperation with Dr. T. Kline of the Prince William Sound Science Center and is described in detail in his Detailed Project Description. Our own work on this aspect will entail analysis of over 1000 samples collected in the vicinity of marine mammal haulouts and feeding areas. It will also include samples from outside of PWS to provide information of

potential shifts in isotope ratios arising from migrations. Dr. Kline estimates the need for analysis on approximately 2000 samples. Analytical costs for these latter samples are included in the separate proposal by Dr. Kline.

3. A service/research component supplying analytical services for carbon and nitrogen isotope ratios to other PI's involved with EVOS studies. This effort will entail consultation and analysis of selected samples to build upon the data base and to integrate the food web studies into a cohesive picture of the trophic dynamics. This task is anticipated to require approximately 20% of the analytical and research effort and has been embraced enthusiastically by other research components. We already have samples from 9 sea otters and collections of sea birds and prey are currently underway.

The ancillary work from other participants will be coordinated through the UAF Stable Isotope Facility and will consist of approximately 1000 samples. If there appears to be more than 1000 external samples collected, the PI will prioritize samples in consultation with the investigators. All work will be performed cooperatively and the data shared as outlined in the Detailed Project Descriptions of the cooperating studies.

D. Completion Date

This project is anticipated to be complete in 1998. The service aspects of the mass spectrometry for isotope ratios may continue beyond that date if demand warrants.

COMMUNITY INVOLVEMENT

The 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 USFWS has collected seal whiskers and tissues for this study in the past and we anticipate this assistance will continue.

FY96 BUDGET (thousands)

Personnel	83.7
Travel	11.2
Contractual	5.2
Commodities	6.9
Subtotal	107.0
Indirect Costs	521.4
Total	128.4

PROJECT DESIGN

A. Objectives

The objectives to be completed during the period of this proposal are essentially the same as in our FY95 proposal and include:

- 1. Collect samples of harbor seal vibrissae through continued cooperative work with the Alaska Department of Fish and Game 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 through assistance by ADF&G personnel monitoring harvests and through the efforts of 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.
- 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. Through the use of captive animals being fed known diets, establish the relationships between whisker growth rate and temporal changes and the fractionation factors between the δ^{13} C and δ^{15} N values of diet and consumer.
- 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.

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 as to location in the face. The whisker is then segmented at 2.5 mm intervals with a razor and the subsamples placed in vials for later grinding and mass spectrometry. The subsamples obtained are dried and powdered for homogeneity and the isotope ratios of carbon and nitrogen determined with a Europa 20/20 mass spectrometer system. The sample is flash combusted at high temperature and the nitrogen and carbon dioxide gases separated and purified by gas chromatography. These are subsequently led into the mass spectrometer by capillary and the isotope ratios determined. The analytical replicability for the entire sampling process is better than $\pm 0.05^{\circ}/00$ for both δ^{13} C and δ^{15} N.
- Sampling - The acquisition of samples for isotope analysis 2. 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, the Alaska Dept. of Fish and Game, 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 Sound and over This will allow "within taxa" comparisons to the seasons. 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 US Fish and Wildlife Service, ADF&G, and the EVOS-sponsored efforts having field programs. Our experience in 1994 has already produced a wide variety of samples and there is reason to anticipate that 1995 will be even more productive as the requests for materials are communicated to field researchers. The small amounts of sample required for isotopic analyses means that little effort for preservation or transport is required.

The application of isotope ratio work with marine mammals is relatively new and the technique is still in a process of calibration. We have been offered the opportunity to conduct captive animal experiments at the Memorial University of Newfoundland and the Mystic Aquarium in Connecticut using harp seals and harbor seals. We plan to conduct measurements of whisker growth rates and correlation experiments between seals and diets of known composition. Seal vibrissae will be marked and growth rates measured over the seasonal cycles to determine if physiological effects are translated into differing isotope ratios. This work will comprise only a limited amount of the total effort but will be essential, given this relatively new field of application. This work will be conducted by Ph. D. student Amy Hirons as part of her dissertation program. This project will support travel costs to Memorial University to establish experiment protocols and to acquire data and information from cooperating investigators.

Tissues samples for analysis from cooperating investigators will be supplied to the P.I. 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 upon the difficulties involved. Similarly, glass fiber filtered samples will be charged at double the normal sample 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.

Synthesis of data - - The plots of isotope ratios of carbon 3. 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, 1993-4 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 ADF&G personnel, the 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 off the mark. Although a complex ecosystem such as Prince William Sound with strong 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.

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C. Contracts and Other Agency Assistance

None

D. 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 in St. John's Newfoundland and at the Mystic Aquarium, Mystic Connecticut. The benefits of this project will be realized throughout the PWS and will be applicable to other areas of the state.

SCHEDULE

A. Measurable Project Tasks for FY96

1 Oct. 95 - Feb. 1996:	Prepare and analyze isotope ratio samples collected in 1994-1995.
15 Feb 31 March 1996:	Synthesis and coordination for sampling in 1996, Annual report on FY96 (and prior) work.
Apr. 1996 - Aug. 1996:	Field work and sampling, captive animal experiments.
Aug. 1996 - Sep. 1996:	Analysis of samples Post field analysis and planning for 1997 zData synthesis, identification of gaps.

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 will be attained as follows:

<u>Captive animal studies</u> of vibrissae growth rates and dietary effects on stable isotope ratios --- Now underway and completion anticipated in late 1996. <u>Field collections of prey species over the geographic region,</u> <u>collections of whiskers and tissues from harbor seals</u> -- Currently underway. Will continue through FY97, but will be more directed toward the end of the study as we fill data gaps.

<u>Stable isotope analyses</u> -- The laboratory work associated with the preparation of samples and the 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 FY96 and FY97.

<u>Modeling and synthesis of results</u> will occur over the entire project in an iterative process with the emphasis building in FY97 and continuing until the conclusion of the project.

<u>Assistance to other investigators</u> -- This aspect is now underway and will continue throughout the project. It is anticipated that the maximum interaction will occur during FY97 and FY97. Synthesis and interpretation of isotope ratio data will be ongoing.

Project milestones and reporting periods:

Oct.	1996 - 1	Feb. 199'	7:	Analysis of 1996 field season samples Preparation of journal manuscripts.
Mar.	1997 - 2	Apr. 199'	7:	Preparation for field, continue analyses Annual Report.
Apr.	1997 - 1	Aug. 1991	7:	Field work, continued analytical work.
Sep.	1997 - I	Dec. 1997	7:	Analytical work, synthesis and completion of captive animal expts.
Jan.	1998 - N	Mar. 1998	B:	Final report, synthesis meetings, manuscript preparation.

C. Project Reports

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

Annual Reports:

These reports will detail progress and preliminary findings and notable achievements. These are anticipated for the ends of FY96 and FY97.

Final Report:

A final Report will be provided. Technical results in these reports will be shared with EVOS collaborators. Thus they will be apprised of the development of the stable isotope methodology and the interpretation of the results. The PI's will provide expertise in interpretation of isotope results in other projects for which the isotope techniques are only a minor portion of the scientific effort. The final reports of the PI's' 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 upon the scientific. findings. These publications will generated by the PI's 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 P.I. and graduate student Amy Hirons.

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 occurthrough 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.

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 with the Alaska Department of Fish and Game, 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 the 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 is supported by the North Pacific Universities Consortium and 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.

ENVIRONMENTAL COMPLIANCE

The sampling and use 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 will be reviewed for renewal in FY97.

PERSONNEL

Dr. D.M. Schell has been involved in stable isotope studies for over 25 years. His work has involved both 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 continued to be upheld and have provided insight into bowhead whale natural history that was unattainable by other techniques.

As P.I., Dr. D.M. Schell will oversee the Quality Assurance/Quality Control aspects of this project. We have established protocols for sampling and our working standards are cross-calibrated with other nationally recognized laboratories. Primary standards are from the National Technical Standards Service. Our mass spectrometer technician has been well-trained and has over eight years experience on three mass spectrometers.

Dr. Schell oversees the Stable Isotope Ratio Mass Spectrometry Facility on the UAF 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. Machine operations are the responsibility of Norma Haubenstock, technician, who has over eight years experience in the laboratory. We have budgeted additional funds for an assistant to prepare samples, load and operate the automated system and to aid in data processing and archiving for all users.

Donald M. Schell Professor, Marine Science P.O. Box 757220, University of Alaska Fairbanks, Fairbanks, AK 99775-7220 Phone: (907) 474-7115 FAX: (907) 474-7204 E-mail: ffdms1@ims.alaska.edu

Date Prepared

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

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Budget Category:	FFY 1995	FFY 1996						
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Personnel		\$6.3						
Travel		\$0.0						
Contractual		\$133.8						
Commodities		\$0.0						
Equipment		\$0.0		LONG F	RANGE FUNDIN	IG REQUIREME	NTS	
Subtotal	\$0.0	\$140.1	Estimated	Estimated	Estimated	Estimated	Estimated	Estimated
General Administration		\$10.3	FFY 1997	FFY 1998	FFY 1999	FFY 2000	FFY 2001	FFY 2002
Project Total	\$0.0	\$150.4	\$148.0	\$127.0	\$0.0	\$0.0	90.0	\$0.0
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Full-time Equivalents (FFE)		0.1						
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	Project Number: 96170 Project Title: Isotope Ratio Studies of Marine Mammals in Prince William						FORM 3A	
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# 1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET October 1, 1995 - September 30, 1996

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		Subtotal		1.0	6,333	0	
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1 I		Project Number: 96170					Personnel
	1996 Project Title: Isotope Ratio Studies of Marine Mammals in Prince William						
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		Agency: AK Dept. of Fish & Game				L	DETAIL
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# 1996 EXXON VALDEZ TRUST OUNCIL PROJECT BUDGET October 1, 1995 - September 30, 1996

Contractual Costs:				Proposed
Description				FFY 1996
RSA issued to	plan, implemen	it and interpret results of stable isotope study		133.8
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When a non-trustee	organization is	used, the form 4A is required. Contractual	Total	\$133.8
Commodities Costs:	:			Proposed
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1330		Project Title: Isotope Ratio Studies of Marine Mammals in Prince William	Co	mmodities
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New Equipment Pu	rchases;		Number	Unit	Proposed
Description			of Units	Price	FFY 1996
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Those purchases a	ssociated with replaced	ment equipment should be indicated by placement of an R.	New E	uipment Total	\$0,0
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1996 EXXON VALDEZ TRUS COUNCIL PROJECT BUDGET October 1, 1995 - September 30, 1996

Budget Category:	Authorized FFY 1995	Proposed FFY 1996						
Personnel		\$83.7						
Trave		. \$11.2						
Contractual		\$5.2						
Commodities		\$6.9				S		
Equipment		\$0.0		LONG	RANGE FUNDI	NG REQUIREMI	ENTS	
Subtotal	\$0.0	\$107.0	Estimated	Estimated	Estimated	Estimated	Estimated	Estimated
Indirect		\$26.8	FFY 1997	FFY 1998	FFY 1999	FFY 2000	FFY 2001	FFY 2002
Project Total	\$0.0	\$133.8	\$130.0	\$110.0	\$0.0	\$0.0	\$0.0	\$0.0
Full-time Equivalents (FTE)		1.3						
			Dollar amount	ts are shown in	thousands of c	Jollars.		· · · · · · · · · · · · · · · · · · ·
Other Resources							×.	
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 1996
 Project Number: 96170
 FORM 4A

 Project Title: Isotope Ratio Studies of Marine Mammals in Prince William
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# 1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET October 1, 1995 - September 30, 1996

Pers	nnel Costs:			Months	Monthly		Pronceed
	Name	Position Description		Budgeted	Costs	Overtime	FFY 1996
22512	Schell, D.	Principal Investigator		3.0	11.564		34.7
	Haubenstock, N.	Technician		4.0	4.583		18.3
	Barnett, B.	Laboratory Asst.		8.0	3.843		30.7
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(\$10) 51	Description		Price	Inps	Days	Per Diem	FFY 1996
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**JUNCIL PROJECT BUDGET** September 30, 1996

October	1.	1995	-	September	30,	1996
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	Proposed
Description	FFY 1996
Communications Maintenance visit, mass spectrometry factory service Shipping, expediting, Cordova-Fairbanks	0.7 4.0 • 0.5
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Contractual Total	\$5.2
Commodities Costs:	Proposed
Description	FFY 1996
Mass spectrometry gases, glassware Chemicals, lab glassware, expendables Data management software Field gear, ring nets, shipping containers	3.5 1.4 0.8 1.2
Commodities Total	\$6.9
1996 Project Number: 96170 Project Title: Isotope Ratio Studies of Marine Mammals in Prince William Sound Name: University of Alaska	RM 4B ractual & modities ETAIL

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

New	Equipment Purchases:		Number	Unit	Proposed
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## **Remote Video System Seabird Monitoring Project**

Project Number: (	94175
Restoration Category:	Monitoring
Proposer:	DOI-FWS (Alaska Maritime National Wildlife Refuge)
Lead Trustee Agency:	USFWS
Cooperating Agency:	None
Duration:	2 Years (FY 96 and FY 97)
Cost FY 96:	\$38,700
Cost FY 97:	\$13,100
Geographic Area:	Field work will be conducted at East Amatuli Island and East Amatuli Light Rock in the Barren Islands, Alaska
Injured Resource/Service:	Common murres

## ABSTRACT

The project is designed to test the ability of a robotically controlled video monitoring system to remotely collect real-time productivity, nesting chronology, adult time budget, and chick feeding rate data on common murres and other seabirds (e.g., black-legged kittiwakes) more accurately and at lower costs than current methods allow at colonies with difficult access. The proposal is based on a prototype system that was designed and successfully tested for basic functions in Kachemak Bay and the Barren Islands in FY 94. Data will be collected both remotely and manually on the same sets of plots at the same study site using the same basic methods in conjunction with the APEX Barren Islands Seabird Study (Project 96163J), and the remotely and manually collected data will be tested for significant differences.

## INTRODUCTION

This is a proposal to field test a robotically controlled video monitoring system that can remotely collect real-time productivity, nesting chronology, adult time budget, and chick feeding rate data on common murres and other seabirds (e.g., black-legged kittiwakes). The proposed work is based on a prototype 1-camera system that was designed and successfully tested in Kachemak Bay and the Barren Islands in FY 94. Preliminary Phase I field trials of the prototype unit focused on testing the basic design and function of the equipment in actual field situations. Because all of the trials were successful, we are requesting support to field test an upgraded state-of-the-art version of the video monitoring system during FY 96.

These Phase II tests are designed to evaluate the system's ability to collect nesting chronology, productivity, and chick-feeding rate information on nesting murres and kittiwakes more accurately and at lower costs than current methods allow at colonies with difficult access. This will be accomplished by operating the video equipment at the same time personnel working on the APEX Barren Islands Seabird Study collect the same types of data manually on the East Amatuli Island - Light Rock study plots (the Barren Islands Seabird Study was funded in FY 95 as APEX Project 96163J, and these studies will be conducted again in FY 96, if the APEX program is reauthorized after the FY 95 field season). Information on Phase I tests, new system refinements, and estimated costs for the proposed Remote Video Seabird Monitoring System Study are provided below.

## NEED FOR THE PROJECT

## A. Statement of Problem

Currently, nesting chronology, productivity, chick-feeding rate, and adult time budget data for many species of cliff-nesting seabirds, including common murres, are collected by physically visiting observations posts and viewing nest sites on study plots on a regular schedule (usually every 2-3 days). This method of obtaining information can be labor intensive and costly at colonies with difficult access, because several people may be needed to collect the data. This method can also be dangerous, because at many locations observers have to boat to study sites, land on rock shelves or boulder beaches, and climb steep unstable slopes to reach observations posts many times during the nesting season (e.g., Nord Island in the Barren Islands). Furthermore, this method of collecting information is highly weather dependent, and unfavorable boating conditions at critical times during the breeding season (e.g., egg-laying, chick-hatching) can degrade or destroy the value of the data sets (i.e., if gaps become too large between observations to calculate hatching dates). To help solve these problems, we are proposing to test a new version of the prototype video system in the Barren Islands during FY 96 to assess the effectiveness of this equipment in seabird monitoring studies.

Although total populations of common murres nesting at the Barren Islands colonies have not changed significantly in size over the 6-year period following the T/V *Exxon Valdez* oil spill, numbers of chicks per adult and productivity values were normal during 1992-1994 and 1993-1994, respectively (Dragoo *et al.* 1994; Roseneau *et al.* 1995; Roseneau *et al.*, unpubl. data). However, productivity information is still needed at these colonies to meet restoration monitoring goals, and these and several other types of data are needed for the Trustee Council APEX projects to help describe relationships between seabirds and forage fish, and for energetics studies (e.g., nesting chronology, chick-feeding rates, and time budget indices of adults). Furthermore, these types of data may also be needed in coming years for long-term research projects.

## B. Rationale

The project is needed because if the video monitoring system passes Phase II testing, it will give the Trustee Council the ability to collect information on timing of nesting events, productivity, chick-feeding rates, and time budgets of adults at colonies with difficult access (e.g., Nord Island, Chiswell Islands) more efficiently and at lower costs, compared to current programs. These types of data are needed to help meet common murre restoration monitoring goals. They are also required for studies of relationships between seabirds, forage fish, and environmental variables (e.g., water temperatures), and

energetics studies of seabirds. In addition to reducing overall costs of collecting these types of information, the video monitoring system has high potential to markedly improve data quality, because observers will be able to collect information at optimum times of day on a much more regular basis (i.e., collecting data via the system will be much less weather dependent than visiting study plots by boat to collect it manually). Because the monitoring system can be easily modified to simultaneously collect and transmit other types of valuable information in conjunction with the video signal at relatively low cost (e.g., air and water temperatures, wind speeds), it also has high potential for use during more complicated studies assessing effects of environmental factors on nesting seabirds. Also, if the video system passes the Phase II tests, it may be useful to other types of ongoing and potential Trustee Council restoration work (e.g., monitoring use or behavior at marine mammal haul-outs and pupping areas) and similar research projects in other regions of Alaska and the world.

## C. Summary of Major Hypotheses and Objectives

The project objective is to test the abilities of a robotically controlled, high resolution Hi-8 mm video monitoring system to collect productivity, nesting chronology, chick feeding rate, and adult time budget indices data on common murres and black-legged kittiwakes more accurately and at lower costs than current more labor intensive manual methods allow at difficult to access colonies. The objective will be met by operating the video equipment at the same time observers collect the same types of information manually on the East Amatuli Island - Light Rock study plots, and testing the video and manually obtained information to see if there are significant differences between data sets.

## D. Completion Date

The proposed project will be completed in FY 97 (i.e., field work will be conducted in FY 96 and a report summarizing study results will be submitted to the Chief Scientist on May 15, 1997).

## **COMMUNITY INVOLVEMENT**

A short article describing the FY 96 Remote Video Monitoring System Study will be prepared for the Trustee Council newsletter after the Phase II tests are complete. Photographs showing operation of the system will also be provided to Trustee Council staff for public viewing, along with a video tape that will describe the equipment and show it in use at the East Amatuli Island - Light Rock seabird colony. The results of the study will be available to the public in Homer and Anchorage, and information from the project will be presented to the public during Trustee Council-sponsored workshops in 1996-1997.

## FY 96 BUDGET

Cost breakdowns for the FY 96 Remote Video Monitoring System Study are shown on Forms 3A and 3B, and are summarized below. Total estimated costs for FY 96 are \$38,700.00, and estimated FY 97 costs for producing a report summarizing system performance are \$13,100.00. Renting primary components of the video system was chosen as the best alternative for conducting Phase II tests, because purchasing equipment would be more expensive. Although we have considerable faith in the system based on Phase I experiments, we believe the equipment should be thoroughly tested before any commitments are made to purchase it. Rental costs are shown in the Equipment section of the budget

(see Form 3B). These rental costs total \$19,800.00 and should not be construed as new equipment purchases.

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## **PROJECT DESIGN**

## A. Objectives

The overall project objective is to test the abilities of a robotically controlled, Hi-8 video monitoring system to collect productivity, nesting chronology, chick feeding rate, and adult time budget indices data on common murres and black-legged kittiwakes more accurately and at less expense than current more labor intensive manual methods allow at colonies with difficult access. The objective will be met by:

- 1. Statistically comparing manual and remote methods of data collection on the East Amatuli Island Light Rock study plots.
- 2. Remotely collecting certain types of data (e.g., productivity information) at optimum times of day and comparing this information with the same types of data collected at other times of day.

## B. Methods

During August 1994, we had the opportunity to field test a video monitoring system that demonstrated potential to collect a variety of data on nesting seabirds A robotically-controlled 8 mm video camera connected to a microwave video transmitter was placed on Nord Island near the observation post historically used to monitor productivity of common murres. During a series of tests, all basic camera functions (e.g., zoom in/out, focus) and a wide range of camera movements (both horizontal and vertical) were easily controlled from line-of-sight distances up to 1 km away. The camera was successfully turned on and off remotely from the Ushagat Island field camp, 6.4 km away, and from other line-of-sight points as far as 11.3 km away (e.g., from a vessel stationed opposite the north end of East Amatuli Island). Images of murres on the productivity plots were also successfully transmitted over these same distances to a video monitor equipped with a microwave receiver. Although neither the camera nor the monitor were high resolution units, the images were clear enough to easily count both adults and chicks on the Nord Island plots. Indeed, on 1 day when seas were too rough to land at Nord Island, observers stationed on a vessel 1 km away used the system to count adults and then zoom in and out and move the camera to check their respective nest sites one at a time for chicks.

Based on the success of the FY 94 tests, we are proposing to test the ability of a refined Hi-8 video monitoring system to collect productivity, nesting chronology, chick feeding rate, and adult time budget data in the Barren Islands during FY 96. The refined equipment is capable of transmitting and receiving clear, crisp video images and camera control signals over line-of-sight distances of 10 km (this distance was tested in Kachemak Bay in late 1994). It can also operate over longer line-of-sight distances and in situations that do not have line-of-sight conditions using relatively low-cost repeater stations.

We propose to use the remotely controlled video monitoring system to collect real-time nesting chronology and productivity data at the East Amatuli Island - Light Rock murre and kittiwake study plots by the same basic methods that observers will use when they physically visit the plots. As during the FY 93 and FY 94 common murre restoration studies (Projects 93049 and 94039), nest sites will be mapped and viewed on a regular schedule (every 2-3 days) to determine presence or absence of eggs and chicks. Incubation and brooding postures of adults will be used to help determine if eggs or chicks are present. The primary difference will be that some observers will view nest sites on a high resolution monitor located several kilometers away instead of viewing them from an observation post with binoculars and spotting scopes. Personnel using the video monitoring system will collect nesting chronology and productivity data at the same times of day information is obtained manually, and also at optimum times of day when fewer birds are present to allow efficiency and accuracy comparisons to be made between data sets. The video system will also be used to collect data on chick feeding rates and time-budget indices of adults for comparison with manually collected information.

The study plot portion of the video system will consist of 2 small robotically controlled, weatherproof, CCD color video cameras connected to a microwave video transmitter and control signal receiver unit. The cameras will be equipped with low-lux telephoto lenses, and attached robotic units will provide a full range of camera movements and lens control at the required distances. The equipment will be powered by self-sustaining power supplies consisting of 12V batteries and solar panels similar to units that we have already successfully used to power time-lapse video cameras for long periods of time. Images will be transmitted from the cameras to a high resolution monitor and Hi-8 video recorder/editor at Amatuli Cove Camp via 2 small microwave - VHF repeater stations mounted on top of East Amatuli Light Rock and Valley Rise (see Fig. 1). Signals controlling camera functions (e.g., sleep/wake, zoom in/out, right/left/up/down) will be transmitted back to the cameras and robotic units through the same relay stations (the repeater units are necessary, because transmitting images and camera control signals requires line-of-sight conditions).

Each of the robotically controlled cameras will be set up to view 5 of the 10 previously studied East Amatuli Island - Light Rock productivity. The cameras will function one-at-a-time (i.e., the system is designed to switch between cameras to minimize costs of transmitting and receiving video and control signals). Telephoto zoom lenses will be used to provide both distant views of plots suitable for counting adults and detailed close-up views of individual nest sites for determining presence or absence of eggs and chicks. The lenses will also have low-lux capability that will allow clear images of birds to be obtained during low light conditions (e.g., during late evening hours in late August - early September).

The video system will be installed in mid-June, at the same time Amatuli Cove Camp is mobilized. Setting up and testing the system will require 5-6 days. The repeater stations and camera units will be removed when the field camp is demobilized in early September. Equipment removal will only require about 1 day. Productivity and nesting chronology data will be analyzed by the same methods described by Roseneau *et al.* (1995), and analyses of chick feeding rate and time budget data will follow methods listed by Roseneau (1995). During data analyses, plots will be treated as sample units, and 2-tailed *t*-tests will be used to check the manually and remotely collected data sets for significant differences (significance level 0.05).

## C. Contracts and Other Agency Assistance

A contract will be required for technical assistance during installation of the video monitoring system, because technical expertise is required to assemble and install the Phase II camera robotic systems and repeater stations, and make adjustments to the equipment during the experiments. The contract will also cover maintenance of the equipment during the study, and training of personnel to operate, adjust, and maintain the equipment.

## D. Location

The FY96 work will be conducted at East Amatuli Island in the Barren Islands, about 75 km southsouthwest of Homer, Alaska between the Kenai Peninsula and the Kodiak archipelago. The project has potential to benefit studies of seabirds and marine mammals in the Gulf of Alaska, including the spill zone, and throughout the remainder of Alaska and elsewhere. No communities will be affected by the study.

## SCHEDULE

<b>A</b> .	Measurable	Project	Tasks	for	FY	96
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January 1 - March 31:	Arrange for equipment rental, technical assistance contract, and integration with other studies
April 1 - May 15:	Arrange other logistical needs (transportation to study area)
May 16 - June 14:	Prepare equipment for transport to Barren Islands study area
June 15:	Transport equipment and technical assistant to Amatuli Cove Camp
June 16 - 25:	Set up equipment at East Amatuli Island study plots
June 26 - September 7:	Collect data.
September 8 - 12:	Remove equipment from study plots and return it to Homer
September 15 - 30:	Review and organize data

January 1 - May 1, 1997: Prepare report and respond to in-house reviewer comments

May 15, 1997: Submit final report to Chief Scientist for peer review

## B. Project Milestones and Endpoints

Data collection portions of the project objectives listed in Part A of the Project Design section of this proposal will be met by September 1, 1996. Data analyses portions of these objectives will be met by January 1, 1997. The project will be complete when the final report is submitted on May 15, 1997.

## C. Project Reports

The proposed study is not a multi year project that will require data collection beyond the FY 96 field season. Therefore, the final report will be submitted on May 15, 1997.

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

The proposed Remote Video Monitoring System Study is highly coordinated with, and contingent upon, the FY 96 APEX Barrens Islands Seabird Study (Project 96163J; if the FY 96 Barren Islands Seabird Study is canceled, the proposed work cannot be done because it will be too costly). All logistical needs, including boating equipment, camping supplies, transportation to and from the Barren Islands, and transportation within the study area will be provided by APEX Project 96163J. Also, APEX Project 96163J personnel will operate the video system after it is set up and use it to collect the data that will be compared with the information that they will obtain manually on the study plots. The proposed study is also coordinated and integrated with the National Biological Service - Minerals Management Service Kachemak Bay - lower Cook Inlet seabird ecosystem study that will be initiated by J. Piatt, NBS, in FY 95 and continued in FY 96. Piatt, is currently contributing \$15.0K to the FY 95 APEX Barren Islands Seabird Study, and he has agreed to provide \$15.0K to help fund the FY 96 portion of this project and help defray some of the logistical costs that will, in turn, help reduce costs of the proposed Remote Video Monitoring System Study (Piatt is interested in the outcome of the video monitoring experiments because the system could make it easier to work at some of his other Gulf of Alaska study sites). The Alaska Maritime National Wildlife Refuge (AMNWR) will contribute some key pieces of equipment to the proposed Phase II effort to help cut overall video system rental costs (21-inch high resolution color monitor; Hi-8 video recorder/editor; video controller unit; all power supplies, including batteries and solar panels).

## **ENVIRONMENTAL COMPLIANCE**

The proposed project is a non-intrusive study that relies on use of video cameras. No permits are required, and based on review of CEQ regulation 40 CFR 1500-1508, this project has been determined to be categorically exempt from the requirements of NEPA, in accordance with 40 CFR 1508.4.

## PERSONNEL

The project manager, project leader, and field team leader are well qualified to undertake the proposed FY 96 study. Brief resumes of these key personnel are provided here.

## A. Project Manager - G. Vernon Byrd

Vernon Byrd received a B.S. degree in wildlife management from the University of Georgia in 1968, did post-graduate studies in wildlife biology at the University of Alaska-Fairbanks in 1975, and completed a M.S. degree in wildlife resources management at the University of Idaho in 1989. His thesis, entitled "Seabirds in the Pribilof Islands, Alaska: Trends and monitoring methods", explored statistical procedures for analyzing kittiwake (*Rissa* spp.) and murre (*Uria spp.*) population data. Mr. Byrd has worked for the U.S. Fish and Wildlife Service for over 20 years, focusing on studies of marine birds in Alaska and Hawaii. His major interests center around monitoring long-term trends in seabird populations, including numbers of birds and reproductive performance at colonies. He has worked at murre colonies in the Aleutian Islands, the Bering and Chukchi seas, and western Gulf of Alaska. Mr. Byrd was a coauthor of the final *T/V Exxon Valdez* oil spill damage assessment report for murres. Also, he was project manager of the 1993 and 1994 common murre restoration monitoring studies (Projects 93049 and 94039, respectively). Mr. Byrd has authored over 45 scientific papers and 50 U.S. Fish and Wildlife Service reports on field studies, and has made about 20 presentations on seabirds at scientific meetings. Mr. Byrd is the supervisory wildlife biologist at the Alaska Maritime National Wildlife Refuge, the premier seabird nesting area in the national public land system.

## Selected Publications

- Byrd, G.V., E.C. Murphy, G.W. Kaiser, A.J. Kondratyev, and Y.V. Shibaev. (In press). Status and ecology of offshore fish-feeding alcids (murres and puffins) in the North Pacific Ocean.
   Proceedings of "Symposium on the Status, Ecology, and Conservation of Marine Birds of the Temperate North Pacific". Canadian Wildlife Service, Ottawa.
- Byrd, G.V., J.L. Sincock, T.C. Telfer, D.I. Moriarty, and B.G. Brady. 1984. A cross-fostering experiment with Newell's race of Manx shearwater. J. Wildl. Manage. 48:163-168.
- Byrd, G.V., and J.C. Williams. Whiskered Auklet. 1993. A chapter describing the biology of the species *in* The birds of North America 76 (A. Poole and F. Gill, eds.). The Academy of Natural Sciences of Philadelphia. 12 pp.
- Byrd, G.V., and J.C. Williams. Red-legged Kittiwake. 1993. A chapter describing the biology of the species *in* The birds of North America 76 (A. Poole and F. Gill, eds.). The Academy of Natural Sciences of Philadelphia. 12 pp.
- Day, R.H. and G.V. Byrd. 1989. Food habits of the whiskered auklet at Buldir Island, Alaska. Condor 91:65-72.
- Springer, A.M. and G.V. Byrd. 1989. Seabird dependence on walleye pollock in the southeastern Bering Sea. Pages 667-677 in Proceedings of the International Symposium on the Biology and Management of Walleye Pollock. Alaska Sea Grant Rep. No. 89-1, Univ. of Alaska-Fairbanks.

#### B. Project Leader - David G. Roseneau

David Roseneau received his B.S. degree in wildlife management and M.S. degree in biology from the University of Alaska - Fairbanks in 1967 and 1972, respectively. His thesis research was on the numbers and distribution of gyrfalcons, Falco rusticolus on the Seward Peninsula, Alaska. He joined the U.S. Fish and Wildlife Service in January 1993 and was project leader of common murre restoration monitoring Projects No. 93049 and 94039 in the Barren Islands during 1993 and 1994. Mr. Roseneau is also principal investigator of the 1995 APEX Barren Islands Sea Bird Study (Project 95163J). Prior to 1993, he was a consulting biologist for 20 years, and he has conducted and managed marine bird, raptor, and large mammal projects in Alaska and Canada for government agencies and private-sector clients. Mr. Roseneau has been involved in several large-scale murre (Uria spp.) population monitoring projects. During 1976-1983, as co-principal investigator of NOAA/OCSEAP Research Unit 460, he conducted monitoring studies of murres and black-legged kittiwakes (Rissa tridactyla) at capes Lisburne, Lewis, and Thompson in the Chukchi Sea, and St. Lawrence, St. Matthew, and Hall islands in the Bering Sea. He also studied auklets (Aethia spp.) at St. Lawrence and St. Matthew islands, and participated in murre and kittiwake projects at Bluff in Norton Sound. In 1984-1986, he participated in follow-up studies of murres and kittiwakes in the northeastern Chukchi Sea, and during 1987-1988 and 1991-1992, he helped conduct additional murre and kittiwake work at capes Lisburne and Thompson, and at Chamisso and Puffin islands. Mr. Roseneau is experienced in collecting and analyzing data on numbers, productivity, and food habits of seabirds; relating trends in numbers and productivity to changes in food webs and environmental parameters (e.g., air and sea temperatures, current patterns); and assessing potential impacts of petroleum exploration and development on nesting and foraging marine birds. He has broad knowledge of rock climbing techniques and has operated inflatable rafts and other outboard-powered boats in the Bering, Chukchi, and Beaufort seas and on various Alaskan rivers in excess of 2,800 hrs. Mr. Roseneau has also accrued several hundred additional hours operating time in small boats and larger, more powerful vessels (e.g. 25 ft, 300-400 hp HydroSports and Boston Whalers) in Kachemak Bay, Prince William Sound, and Kenai Peninsula and Barren Island waters. During his career, Mr. Roseneau has authored and co-authored 70 reports and publications, including 23 on Alaskan seabirds.

#### Selected Seabird Publications

- Murphy, E.C., A.M. Springer, and D.G. Roseneau. 1986. Population status of *Uria aalge* at a colony in western Alaska: results and simulations. Ibis 128: 348-363.
- Murphy, E.C., A.M. Springer, and D.G. Roseneau. 1991. High annual variability in reproductive success of kittiwakes (*Rissa tridactyla* L.) at a colony in western Alaska. J. Anim. Ecol. 60: 515-534.
- Murphy, E.C., D.G. Roseneau, and P.J. Bente. 1984. An inland nest record for the Kittlitz's murrelet. Condor 86: 218.
- Springer, A.M., E.C. Murphy, D.G. Roseneau, C.P. McRoy, and B.A. Cooper. 1987. Paradox of pelagic food webs in the northern Bering Sea - I. Seabird food habits. Cont. Shelf Res. 7: 895-911.

- Springer, A.M. and D.G. Roseneau. 1985. Copepod-based food webs: auklets and oceanography in the Bering Sea. Marine Ecol. Prog. Ser. 21: 229-237.
- Springer, A.M., D.G. Roseneau, D.S. Lloyd, C.P. McRoy, and E.C. Murphy. 1986. Seabird responses to fluctuating prey availability in the eastern Bering Sea. Marine Ecol. Prog. Ser. 32: 1-12.
- Springer, A.M., D.G. Roseneau, E.C. Murphy, and M.I. Springer. 1984. Environmental controls of marine food webs: food habits of seabirds in the eastern Chukchi Sea. Can. J. Fish Aquat. Sci. 41: 1202-1215.

## C. Field Team Leader - Arthur B. Kettle

Arthur Kettle received his B.A. degree in Human Ecology from the College of the Atlantic in 1984. Since that time, he has participated in several large-scale seabird projects at remote locations. He joined the U.S. Fish and Wildlife Service in May 1993, and is currently the field team leader for the APEX Barren Islands Seabird Study (Project 95163J). In that capacity, Mr. Kettle is responsible for logistics at the Amatuli Cove camp. He is also responsible for ensuring that the data are collected according to the study design. Mr. Kettle was in charge of the East Amatuli Island camp during the 1993 and 1994 common murre restoration monitoring projects (Projects 93049 and 94039). During these studies, his broad knowledge of boat-mooring systems and technical rock climbing techniques allowed him to safely collect productivity and chronology data from a series of study plots he established on East Amatuli Island in 1993 (a difficult technical task not accomplished during any previous pre- or postspill study). Mr. Kettle also collected murre productivity and nesting chronology data at Light Rock during Exxon-sponsored University of Washington Barren Islands studies in 1990-1992. Besides his murre work, he also participated in large-scale University of Washington studies of magellanic penguins (Spheniscus magellanicus) in Argentina in 1987-1991, and tufted puffins (Fratercula cirrhata) and forktailed storm-petrels (Oceanodroma furcata) in the Barren Islands during 1990-1992. Mr. Kettle has over 15 years experience safely operating small boats in both north Atlantic and Pacific ocean waters (Maine and Alaska). By September 1995, he will have completed his sixth consecutive field season operating outboard-powered inflatable and ridged-hulled craft in the Barren Islands.

## Selected Publications

Boersma, P.D., J.K. Parrish, and A.B. Kettle. 1993. Common murre abundance, phenology, and productivity on the Barren Islands, Alaska: The *Exxon Valdez* oil spill and long-term environmental change. In BOOK TITLE, ASTM STP 1219._. (eds.); American Society for Testing and Materials, Philadelphia; 1993.

David G. Roseneau, Project Leader DOI-FWS -Alaska Maritime National Wildlife Refuge (U.S. Fish and Wildlife Service) 2355 Kachemak Bay Drive (Suite 101), Homer, Alaska 99603-8021 Telephone (907) 235-6546 Fax (907) 235-7783 e-mail: r7amnwr@mail.fws.gov

<u>H. Venn Byrd</u> G. Vernon Byrd, Project Manager

G. Vernon Byrd, Project Manager DOI-FWS Alaska Maritime National Wildlife Refuge (U.S. Fish and Wildlife Service) 2355 Kachemak Bay Drive (Suite 101), Homer, Alaska 99603-8021 Telephone (907) 235-6546 Fax (907) 235-7783 e-mail: r7amnwr@mail.fws.gov

April 30, 1995

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- Dragoo, D.E., G. V. Byrd, D.G. Roseneau, D.A. Dewhurst, J.A. Cooper, and J.H. McCarthy. 1994. Effects of the *T/V Exxon Valdez* oil spill on murres: A perspective from observations at breeding colonies four years after the spill. Final rept., Restoration Proj. No. 11, U.S. Fish Wildl. Serv., Homer, AK.
- Roseneau, D.G., A.B. Kettle, and G.V. Byrd. 1995. Common murre restoration monitoring in the Barren Islands, Alaska, 1993. Unpubl. final rept. by the Alaska Maritime National Wildlife Refuge, Homer, Alaska for the *Exxon Valdez* Oil Spill Trustee Council, Anchorage, Alaska (Restoration Project 93049). 82 pp.
- Roseneau, D.G. 1995. Barren Islands seabird studies. Pp. 63-69 in D.C. Duffy (ed.), APEX: Apex predator ecosystem experiment in Prince William Sound and the Gulf of Alaska. A proposal to the *Exxon Valdez* Oil Spill Trustees, March 1995. 103 pp.



Figure 1. The FY 96 Remote Video Seabird System Monitoring Project study area in the Barren Islands. Real time productivity, nesting chronology, time budget, and chick feeding rate data will be collected from the Lonesome Cove productivity plots via a high resolution 8 mm video system that will transmit images from two remotely controlled robotic video camera units to the Amatuli Cove Camp via small microwave - VHF repeater stations located at East Amatuli Light Rock and Valley Rise. Signals controlling camera functions (e.g., sleep/wake, zoom in/out, right/left/up/down) will be transmitted from Amatuli Cove Camp back to the cameras via the same repeater link (dashed lines show approximate line-of-sight transmission pathways). Data collected by the video system will be compared with the same types of data collected on the same plots by a team of observers.

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

r	Authorized	Proposed						
Budget Category:	FFY 1995	FFY 1996						
Personnel		\$8.3						
Travel		\$0.0						
Contractual		\$8.3						
Commodities		\$0.3						
Equipment		\$20.0		LONG RA	<b>NGE FUNDIN</b>	IG REQUIREN	MENTS	
Subtotal	\$0.0	\$36.9	Estimated	Estimated	Estimated	Estimated	Estimated	Estimated
General Administration		\$1.8	FFY 1997	FFY 1998	FFY 1999	FFY 2000	FFY 2001	FFY 2002
Project Total	\$0.0	\$38.7	\$13.1					
Full-time Equivalents (FTE)		0.2						
			Dollar amounts	s are shown ir	n thousands of	dollars.		
Other Resources								
Comments:								
The proposed project is closely	tied to and dep	pendent on the	e proposed FY	96 APEX Bar	ren Islands Se	eabird Study (F	Project 96163)	. If this
\$95,900 study is approved, it will	ill supply all log	istical needs a	and most of the	e personnel n	eeded to cond	uct the work.	The exception	is the
Student Conservation Associati	on volunteer.							
Estimated FFY 1997 costs are of	costs for produ	cing a final re	port summarizi	ng results of t	the FFY 1996	field work.		
			- <u>n. 46. 46. 76. 76.</u> 76.				<u>- Angelin (n. 1965)</u>	
							<b></b>	
	Drojoot Nun	abor						FORM 3A
1006			lide a Cueta		Inclusion D			AGENCY
1990	Project Litle: Remote Video System Seabird Monitoring Project							PROJECT
	Agency: D	JI-FWS						DETAIL
Preparedt -							L	
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## 1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET October 1, 1995 - September 30, 1996

Per	sonnel Costs:	GS/Range/	Months	Monthly		Proposed	
PM	Name	Position Description	Step	Budgeted	Costs	Overtime	FFY 1996
1	D. Roseneau	Wildlife Biologist	GS 11/4	1.0	4,300		4.3
	V. Byrd	Supervisory Biologist	GS 12/4	0.5	5,000		2.5
*	C. Berg	EVOS Coordinator/Program Manager	GS 12/4	0.3	5,000		1.5
							0.0
							0.0
							0.0
ĺ							0.0
						(	0.0
							0.0
							0.0
							0.0
							0.0
		Subtotal		1.8	14,300	0	
Tho	se costs associated with pro	gram management should be indicated by	placement of	an *.	Pe	rsonnel Total	\$8.3
Tra	vel Costs:		Ticket	Round	Total	Daily	Proposed
PM	Description		Price	Trips	Days	Per Diem	FFY 1996
1		None Required					0.0
							0.0
							0.0
							0.0
							0.0
							0.0
1							0.0
							0.0
							0.0
							0.0
							0.0
L						The state	0.0
Ino	se costs associated with pro	gram management should be indicated by	placement of	an ".		Travel Total	\$0.0
		r	······			r	
		Drojost Number				F	ORM 3B
Project Number:					Personnel		
	1990	Project Title: Remote Video Syste	m Seabird N	Monitoring Pi	roject		& Travel
		Agency: DOI-FWS					
	I					L	

## 1996 EXXON VALDEZ TRUS1 COUNCIL PROJECT BUDGET October 1, 1995 - September 30, 1996

Contractual Costs:			Proposed
Description			FFY 1996
Technical Assistan To p the r of pe Student Conservat	ice Contract rovide expert help during assembly of the camera robotic systems and during installation of the nicrowave/VHF repeater stations. The contract will also cover maintainence of the equipment ersonnel to operate and adjust the equipment. The contract will run 2 months @ \$2,435/mo = i tion Association Volunteer Contract	ese units and and training \$4,870.	4.9
One each	SCA volunteer will be used to work with the cameras; cost is \$3,120 for 12 wk contract plus \$ additional week. Need this volunteer for 14 weeks: basic cost \$3,120 + \$112(2) = \$3,350	112/wk for	5.4
When a non-trustee org	panization is used, the form 4A is required. Cor	ntractual Total	\$8.3
Commodities Costs:			Proposed
Description			FFY 1996
	Comn	nodities Total	\$0.3
1996	Project Number: Project Title: Remote Video System Seabird Monitoring Project Agency: DOI-FWS	F( Con Cor E	ORM 3B htractual & nmodities DETAIL

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

New Equipment Purchases:	Number	Unit	Proposed
Description	of Units	Price	FFY 1996
Video System tool kit (new purchase)	1	200	0.2
RENTAL			0.0
Rental of remote video system equipment	1 syste	19,800	19.8
1. Two robotic video camera units with telephoto lenses: 3 mo @ \$1,000/mo/ea = \$6,000			0.0
<ol> <li>Two microwave/VHF repeater units: 3 mo @ \$1,500/mo/ea = \$9,000</li> </ol>			0.0
3. One VHF control signal transmitter : 3 mo @ \$300/mo = \$900			0.0
4. One VHF control signal receiver: 3 mo @ \$300/mo = \$900			0.0
5. One microwave transmitter: 3 mo @ \$500/mo = \$1,500			0.0
6. One microwave receiver: 3 mo @ \$500/mo = \$1,500			0.0
			0.0
			0.0
			0.0
			0.0
Those purchases associated with replacement equipment should be indicated by placement of an R.	New Eq	upment Total	\$20.0
Existing Equipment Usage:		Number	Inventory
Description		of Units	Agency
Inflatable ratt		1	DOI-FWS
Outboard motors		2	DOI-FWS
Climbing equipment		1 Set	DOI-FWS
Solar Panels		2	DOI-FWS
			DOI-FWS
			DOI-FWS
		1	DOI-FWS
		<u> </u>	<u> </u>
		_	001100
Project Number:		F	OKW 3B
1996 Project Title: Remote Video System Seabird Monitoring P	roject		quipment
Agency: DOI-FWS			DETAIL
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## Restoration of Essential Wetland Habitat at San Juan Bay on Montague Island

Project Number: 96	176			
Restoration Category: General Restoration				
Proposer:	USFS			
Lead Trustee Agency:	USFS			
Cooperating Agencies:	None			
Duration:	6 yrs.			
Cost FY 96:	\$67,500			
Cost FY 97:	\$130,350			
Cost FY 98:	\$100,000			
Cost FY 99:	\$50,000			
Cost FY 00:	\$50,000			
Cost FY 01:	\$5,000			
Geographic Area:	Prince William Sound			
Injured Resource/Service	None			

#### ABSTRACT

This project has the potential to create wetland habitats used by waterfowl and anadromous fish impacted by the oil spill in Prince William Sound. A study will be carried out in FY 96 to determine project feasibility from hydrologic, soils, geomorphology, fisheries, wildlife, and engineering perspectives. A detailed project plan will be developed if findings warrant. An environmental analysis will be conducted in FY 97. If the project is implemented, succession will be reversed in the uplifted lake at San Juan Bay on Montague Island. Flooding of the uplifted area will maintain the wetland component. Pools and ponds will be created in riparian and floodplain areas to restore associated aquatic vegetation.

#### INTRODUCTION

Past events associated with the 1964 earthquake drained the 250 ha lake within the San Juan Bay Drainage. Since the uplift, periodic flooding occurs during periods of high, nearly continuous rainfall or during periods of heavy rain combined with snow melt. With this altered water regime, the uplifted lake is undergoing a rapid succession from an estuary/wetland community to a spruce/hemlock community. At the same time, downcutting of the San Juan Creek has changed the character of the stream along a major portion of its course through the former lake bed. Pool habitats important for anadromous fish rearing have been reduced and adjacent sedge meadows are undergoing plant succession to shrub and forest communities. Opportunities exist for long term improvement of PWS waterfowl, furbearer and anadromous fish habitat within the stream and in the adjacent wet meadow zones.

## NEED FOR THE PROJECT

#### A. Statement of Problem

This project has the potential to improve habitat for waterfowl and anadromous fish. This project falls within the category of habitat protection, acquisition, and manipulation of resources. This project will improve or supplement stream and lake habitats.

#### **B.** Rationale

The opportunity exists to create wetland habitat to offset damages to wetland areas that were damaged during the spill and will take a long time to recover. Impounding water in the San Juan drainage will flood areas that were tidal estuaries prior to the 35 foot uplift associated with the 1964 earthquake. Flooding areas with freshwater will reverse succession and create wetland habitat that may be used by waterfowl and salmon.

#### C. Summary of Major Hypotheses and Objectives

A feasibility study will occur in FY 96. If the project is determined to be feasible from hydrologic, soils, geomorphologic, fisheries, wildlife, and engineering perspectives, then the project objectives will be:

- 1. Maintain a wetland component by flooding the uplifted lake bed and reversing succession from a forested habitat type to a grass/sedge community.
- 2. Create pools and ponds in riparian and floodplain areas to restore associated aquatic vegetation.

#### **D.** Completion Date

The feasibility portion of the project will be completed by January 1, 1997. If the manipulative portion is implemented, it will be completed in 1998, and monitoring and other follow-up work will continue another 2 years through fiscal year 2000. Final report would be completed by January 1,

2001.

#### **COMMUNITY INVOLVEMENT**

None.

#### **FY 96 BUDGET**

Personnel	41.9
Travel	6.8
Contractual	7.0
Commondities	5.0
Equipment	0.0
Sub-total	60.7
Gen. Admin.	6.8
Total	67.5

#### **PROJECT DESIGN**

- A. Objectives
- 1. Maintain a wetland component by flooding the uplifted lake bed and reversing succession from a forested habitat type to an early succession grass/sedge community.
- 2. Create pools and ponds in riparian and flood plain areas to restore associated aquatic vegetation.

#### **B.** Methods

This project will be accomplished by the following sequence of events:

1. Determine Feasibility.

This consists of conducting an hydrologic analysis to determine subsurface flow regimes, soils analysis to determine soils types, and a channel morphology analysis. Monthly surveys will determine wildlife and fish use of the area from spring through fall.

2. Inventory Existing Habitat.

Low level aerial photography of San Juan Bay area and vegetation surveys will be used to determine existing plant community type.

3. Design Project.

After the feasibility and inventory studies are completed, vegetation objectives will be established specific to the targeted species. The engineering design will meet those objectives.

4. Conduct Environmental Analysis.

An environmental analysis will be conducted prior to a decision for any action. The scope of the analysis will depend on the results of public scoping and issues developed.

5. Implement Project.

If the decision is made to implement the project, it would take three years to complete.Monitoring.

Monitoring will continue for three years after completion of the project to determine if the vegetation objectives were met.

#### C. Contracts and Other Agency Assistance

A contract will be let for large-scale aerial photography.

#### D. Location

San Juan Bay is located on southwestern Montague Island in Prince William Sound.

#### **SCHEDULE**

#### A. Measurable Project Tasks for FY 96

A report of findings from the feasibility study will be completed for work done in FY 96.

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

The following is the proposed schedule:

January 1 - July 15, 1996:	Hydrologic Analysis
April 1 - October 1, 1996:	Wildlife Surveys
June 1 - 30, 1996:	Engineering Data Collection
June 15 - 30, 1996:	Soils Analysis
July 1 - 15, 1996:	Low aerial Photography
September 1 - 30, 1996:	Project Design
January 1 - April 1, 1997:	Environmental Analysis
1997 - 1998:	Implementation

#### 1997 - 2000: Monitoring

#### C. Project Reports

A feasibility report will be prepared. Annual progress reports will be prepared during project implementation.

## COORDINATION AND INTEGRATION OF RESTORATION EFFORT

Coordination will occur after the feasibility study is completed.

#### ENVIRONMENTAL COMPLIANCE

Given the scope of this project an environmental analysis will be required before a decision is made to implement.

#### PERSONNEL

Personnel needed to determine the feasibility of the project include a GS-11 Hydrologist, GS-11 Soils Scientist, GS-11 Wildlife Biologist, GS-11 Engineer, GS-7 Engineering Technician, GS-7 Wildlife Biologist, and a GS-5 Wildlife Technician.

Ken Holbrook, Project Leader USFS Chugach National Forest 3301 C Street, Suite 300 Anchorage, AK 99503-3998 Phone: (907)271-2819 fax: (907)271-3992

Ray Thompson, Project Manager USFS, Chugach National Forest 3301 C Street, Suite 300 Anchorage, AK 99503-3998 Phone: (907)271-2536 fax: (907)271-3992

# Date prepared

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

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	Authorized	Proposed						
Budget Category:	FFY 1995	FFY 1996						
Personnel		\$41.9						
Travel		\$6.8						
Contractual		\$7.0						
Commodities	· · · · · · · · · · · · · · · · · · ·	\$5.0						
Equipment		\$0.0		LONG F	ANGE FUNDIN	IG REQUIREME	115	
Subtotal	\$0.0	\$60.7	Estimated	Estimated	Estimated	Estimated	Estimated	Estimated
General Administration		\$6.8	FFY 1997	FFY 1998	FFY 1999	FFY 2000	FFY 2001	FFY 2002
Project Total	\$0.0	\$67.5	\$90.5	\$60.0	\$60.0	\$60.0	\$60.0	
Full-time Equivalents (FTE)		0.8						
			Dollar amount	<u>s are shown in</u>	thousands of o	Iollars.		· · · · · · · · · · · · · · · · · · ·
Other Resources	•		•		·			
Comments:								
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	Prince Willia	m Sound						AGENCY
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	Agency: US	<b>r</b> ð						
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1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

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Pers	onnel Costs:		GS/Range/	Months	Monthly		Proposed
PM	Name	Position Description	Step	Budgeted	Costs	Overtime	FFY 1996
	Dave Blanchet	Hydrologist	GS-11/6	1.5	4,400		6.6
	Dan Logan	Wildlife Biologist	Gs-11/6	2.0	4,400		8.8
	Dean Davidson	Soils Scientist	GS-11/6	1.5	4,400		6.6
	Engineer	Engineer	GS-11/6	1.0	4,400		4.4
	Engineering Tech.	Engineering Tech.	GS-7	1.0	3,300		3.3
	Wildlife Tech.	Wildlife Tech.	GS-7	1.0	3,300		3.3
	Wildlife Tech	Wildlife Tech	GS-5	1.0	3,000		3.0
*	R. Thompson	Program Manager	GS13	1.0	5,928		5.9
		· ·					0.0
							0.0
							0.0
							0.0
<u> </u>	·	Subtotal		10.0	33,128	0	
Tho	se costs associated with prog	ram management should be indicated by place	ment of an *.		F	Personnel Total	\$41.9
Trav	el Costs:		Ticket	Round	Total	Daily	Proposed
PM	Description		Price	Trips	Days	Per Diem	FFY 1996
	Air Charter to field site		400	8	40	60	5.6
							0.0
	flights to anchorage		200	2	6	135	1.2
							0.0
							0.0
					,		0.0
					¢		0.0
							0.0
					,		0.0
							0.0
							0.0
		· · ·					0.0
Tho	se costs associated with prog	ram management should be indicated by place	ment of an 🔭			Travel Total	\$6.8
	······································				·		
							FORM 3B
		Project Number:					Personnel
	1996	Project Title: Wetlands Restoration	•				P Traval
		Agency: USES					a Travel
							DETAIL
	2 - ( 4				j	-	5/1/05

5/1/95

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

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Contractual Costs:			Proposed
Description			FFY 1996
Aerial Photo ( Report public	Contract ation/printing		5.0 2.0
		· · ·	
When a non-truste	ee organization is used, the form 4A is required.	Contractual Total	\$7.0
Commodities Cost	IS:		Proposed
Field supplies	<u> </u>		<u> </u>
L		<u>Commodities Total</u>	\$5.0
1996	Project Number: Project Title: Wetlands Restoration Agency: USFS	F Co Cc	FORM 3B ntractual & ommodities DETAIL
	3 of 4		5/1/95

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# 1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

New	v Equipment Purchases:	Number	Unit	Proposed
Desc	cription	of Units	Price	FFY 1996
				0.0
			• •	
				0.0
				0.0
				0.0
			ľ	0.0
	· · · ·			0.0
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
	se purchases associated with replacement equipment should be indicated by placement of an R.	New Ec	upment I otal	\$0.0
Exist	ting Equipment Usage:		Number	Inventory
Desc	cription		of Units	Agency
				i
	,			
		,		
L				
<b></b>				
	Project Number:		F	ORM 3B
	1996 Project Titlle: Wetlands Bestoration		E	quipment
	Agency: USES		ĺ	
			•	

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96177A

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## Cutthroat Trout, Dolly Varden Char Habitat Restoration, Lake Elsner Area

Project Number:	9 <del>6058 - B</del> 467777
Restoration Category:	General Restoration
Proposer:	USFS, Cordova Ranger District
Lead Trustee Agency:	USFS
Duration:	3 years
Cost FY 1996:	<b>\$2</b> 6.6
Cost FY 1997:	Dependent on results of work in FY 1996
Cost FY 1998:	Dependent on work in previous years
Geographic Area:	Prince William Sound, Lake Elsner watershed, east of Cordova
Injured Resource:	Cutthroat trout, Dolly Varden char

## ABSTRACT

Timber harvests in the Lake Elsner watershed, 13 miles east of Cordova, may have affected cuthroat trout and Dolly Varden char habitat. The Cordova Ranger District proposes to work with the Eyak Corporation to survey the area and determine if there are any existing or potential impacts. If problems are identified, plans for restoration projects will be developed.

## INTRODUCTION

In recent years there has been increasing concern over the cutthroat trout and Dolly Varden char populations in the Prince William Sound area. Studies by the Alaska Department of Fish and Game show that these species were adversely affected by the *Exxon Valdez* oil spill (Hepler et al. 1993). Local fishermen have also reported decreased numbers of fish, which has led to sportfishing closures and reduced bag limits throughout the area. It seems that the best long-term method of preserving these stocks is to protect and restore the habitats these species utilize.

At the present time it is not known whether timber harvest in the Lake Elsner watershed has affected their habitat or not. It is known, however, that harvest has occurred around streams and lakes with cutthroat trout and Dolly Varden char populations. The Cordova Ranger District proposes to work with the Eyak

Corporation to identify any areas where restoration projects may be needed. Such work could include rehabilitating stream crossing sites, closing and reseeding roads that are no longer needed, or improving the riparian vegetation. If it is found that there are restoration opportunities, plans for rehabilitation projects can be developed for FY 1997.

The role of the Forest Service would be to take the lead in contacting the Eyak Corporation and other concerned groups in order to develop a plan to survey the watershed and identify any possible impacts to fish habitat. If restoration work is necessary, the Forest Service could help develop proposals, assist with the permit process, and work on the environmental analysis. Since the work would be on Eyak Corporation land, the corporation would be responsible for hiring crews and carrying out the project in FY 1997. The role of the Forest Service at this point would be to provide technical assistance if needed.

## NEED FOR THE PROJECT

## A. Statement of Problem

Studies by Hepler et al. (1993) showed not only that cutthroat trout had been adversely impacted by the *Exxon Valdez* oil spill, but also that many of the populations are quite small, making them more vulnerable to negative effects. The numbers of Dolly Varden char are generally higher, but their survival rates were found to be affected more by the oil spill than the cutthroat trout. Fishermen in the Cordova area have also reported that cutthroat trout populations have declined. It is not known to what degree either species has been affected or if they have made any recovery from the oil spill or other events. However, until more is known, it will be especially important to protect the stocks and habitat that remain and restore those areas which have been degraded.

The area in the Lake Elsner watershed has been logged, but it is not known if the fish habitat has been affected. It is possible that there are no problems. However, if there are areas where erosion from road crossings or other problems exist, it would be best to identify those problems now before they get worse This project would ensure that we are aware of any potential threats to fish habitat and have a plan to restore any degraded habitat .

## B. Rationale

Although this area was not directly affected by the *Exxon Valdez* oil spill, Hepler et al. (1993) found that cutthroat trout and Dolly Varden char can stray considerable distances and move into different watersheds. Thus, it seems judicious to try to protect or restore habitat throughout the Sound. Protection of the habitat is also the best way to help maintain healthy populations and protect them against adverse impacts in the future, which is one of the objectives of the cutthroat trout/Dolly Varden char recovery plan.

## C. Summary of Major Hypotheses and Objectives

The main objectives for FY 1996 are to develop a memorandum of understanding with the Eyak Corporation to conduct a joint survey of the Lake Elsner watershed, identify any problems that may

affect fish habitat, and develop a restoration plan, if necessary. If the Eyak Corporation agrees with this project, restoration work would be carried out in FY 1997 by crews hired by the corporation. At this point it is thought that most of the work would involve removing culverts at stream crossings that are no longer needed, rehabilitating stream banks at those sites, and controlling any erosion that introduce sediment into the streams. Monitoring would occur in FY 1998.

## D. Completion Date

Restoration work should be completed in one field season (FY 1997), and the monitoring should be conducted in FY 1998.

## **COMMUNITY INVOLVEMENT**

The restoration work and hiring of the crews would be conducted by the Eyak Corporation. The crews would most likely be composed of local residents and members of the Eyak tribe. Thus, the local community would be directly involved in the project.

## FY 96 BUDGET

Personnel	20.2
Travel	1.5
Contractual	1.5
Commodities	0.3
Equipment	0.0
Subtotal	23.5
Gen. Admin	3.1
Total	26.6

## **PROJECT DESIGN**

## A. Objectives

The main objective for FY 1996 is to conduct stream surveys and other watershed analyses to identify factors which could have adverse effects on cutthroat trout habitat. It is thought that the most likely problems would be bank erosion at stream crossings or other factors leading to stream sedimentation. If problems are identified, a restoration plan would be developed. In FY 1997 the restoration work would be carried out. In FY 1998 the work would be monitored.

## B. Methods

Streams in logged areas will be identified from recent aerial photographs. Crews will walk up each stream and note the following:

- 1. Presence of suitable spawning or rearing habitat.
- 2. Bank erosion or other conditions which may lead to stream sedimentation.
- 3. Adequacy of riparian vegetation for input of large woody material, shading.
- 4. Presence of excessive slash or other logging debris.
- 5. Other unfavorable conditions.

Crews will also survey the roads to identify conditions which could affect streams located downhill from the roads. This could include:

- 1. Culverts or ditches which concentrate runoff and are causing erosion.
- 2. Destabilized slopes.
- 3. Large bare areas which could be eroded.
- 4. Roads which channel runoff and sediment to the streams.

If problems are identified, the location will be recorded and a brief restoration prescription will be written. Restoration measures could include:

- 1. Stabilizing or protecting eroding banks with rip-rap or log structures.
- 2. Revegetating bare or eroded areas with native plants.
- 3. Removing culverts and rehabilitating stream crossing sites.
- 4. Closing unneeded roads and revegetating.

It should be noted that buffer strips were left around most streams, so the work may be limited to stream crossing sites and other road work.

## C. Contracts and Other Agency Assistance

No contracts would be needed for the actions proposed in FY 1996. Depending on how the Eyak Corporation would like to handle the restoration work, contracts might be needed in FY 1997.

We would coordinate our efforts with the Alaska Department of Fish and Game, Habitat Division and incorporate any suggestions, advice, or ideas they may have. Otherwise, no other agency assistance is needed.

## D. Location
The project area would be in the Lake Elsner watershed, approximately 13 miles east of Cordova, Cordova C-4 quadrangle, T 15S, R 1W, sections 16-21 and 28-33. The benefits of the project would be realized in a widespread area. Since this project will also protect coho and sockeye salmon, commercial and recreational fishers from Cordova and elsewhere will benefit. Other anadromous cutthroat trout or Dolly Varden char populations in Prince William Sound may benefit, depending on the amount of straying. The residents of Cordova would also benefit from the job opportunities created by this project, depending on how the hiring is conducted.

## SCHEDULE

## A. Measurable Project Tasks for FY 96

October to December 1995:Meet with participants, develop MOU.January to March 1996:Hire study crew, develop data sheets, logistics.May to July:Conduct surveys, make initial prescriptions.July:Revisit sites, finalize work plans.August to September:Annual report.

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

- 1. Surveys and prescriptions will be completed by July 1996.
- 2. Restoration work will begin May 1997 and end in September 1997.
- 3. Monitoring will be completed by July 1998.

## C. Project Reports

Since this project does not include lengthy data analysis and the project work will occur in the summer months, annual reports can be completed by the end of each fiscal year.

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

At this point we do not foresee any opportunities for coordination or integration with other agencies or restoration efforts.

Since this project would be on land owned by the Eyak Corporation, it would not be part of the normal Forest Service management responsibility. The Cordova Ranger District is proposing this project to promote the overall health of the cutthroat trout and Dolly Varden char populations in the area and to help maintain angling opportunities on non-Forest Service land to keep fishing pressure dispersed.

## **ENVIRONMENTAL COMPLIANCE**

The proposal for the actual restoration work would have to be reviewed by the State of Alaska Division of Governmental Coordination and the U.S. Army Corps of Engineers. The Corps permit for the project should be covered by the Nationwide Permit Program, permit numbers 13 and 27, which authorize erosion control work and restoration of riparian areas. The U.S. Forest Service could conduct the NEPA review, which would necessitate a biological evaluation, public scoping, and a categorical exclusion.

## PERSONNEL

Project Leader:

Ken Hodges USDA Forest Service, Cordova Ranger District P.O. Box 280, Cordova, AK 99574 (907) 424-7661 (telephone) (907) 424-7214 (Fax)

Responsibilities: Plan surveys, train crews, analyze data, make recommendations for restoration work.

Ken Hodges is a fisheries biologist on the Cordova Ranger District. He has a B.S. degree in fisheries from Humboldt State University. Before coming to the District in 1989, he had worked as a seasonal employee for the Oregon Dept. of Fish and Wildlife and conducted a one-year study on steelhead genetics in Northern California. In Cordova he has worked as a fisheries technician and now as a biologist.

Project Manager:

Ray Thompson USFS Chugach National Forest 3301 C Street, Suite 300 Anchorage, AK 99503 phone (907) 271-2500

Date Prepared

## Literature Cited

Hepler, K.R., P.A. Hansen, and D.R. Bernard. 1993 Impact of oil spilled from the Exxon Valdez on survival and growth of Dolly Varden and cutthroat trout in Prince William Sound, Alaska. Alaska Department of Fish and Game. Anchorage, Alaska.

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

Durdana Ontenna	Authorized	Proposed						
Budget Category:	FFY 1995	FFY 1996						
Personnel		\$20.2						
Travel		\$1.5						
Contractual		\$1.5						
Commodities		\$0.3						
Equipment		\$0.0		LONG F	RANGE FUNDIN	G REQUIREMEN	NTS	<b></b>
Subtotal		\$23.5	Estimated	Estimated	Estimated	Estimated	Estimated	Estimated
General Administration		\$3.1	FFY 1997	FFY 1998	FFY 1999	FFY 2000	FFY 2001	EFY 2002
Project Total		\$26.6	\$0.0	\$0.0	\$0.0	\$0.0	\$0.0	\$0.0
Full-time Equivalents (FTE)		0.4						
			Dollar amount	s are shown in	thousands of d	iollars.		
Other Resources								
			- 01					
1996	Project Num Project Title Elsner Agency: U	iber: <del>96058</del> :Cutthroat Tr SFS		A arden Char H	labitat Restor	ration, Lake		FORM 3A AGENCY PROJECT DETAIL
Prepared: 1	4/28/95 L							5/1/95

# 1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

Pers	onnel Costs:		GS/Range/	Months	Monthly		Proposed
PM	Name	Position Description	Step	Budgeted	Costs	Overtime	FFY 1996
	D.Schmid	Project Leader	GS-11	0.7	4,620	0	3.2
Ì	J. Hodges	Project Support	GS-9	1.0	3,864	0	3.9
	GS-7 -one	Fisheries Tech	GS-7	1.0	3,339	0	3.3
	GS-5-one	Fisheries Tech	GS-5	1.0	2,058	0	2.1
	GS-4-one	Fisheries Tech	GS-4	1.0	1,764	0	1.8
÷	R.Thompson	Program manager	GS-13	1.0	5,928		5.9
							0.0
							0.0
							0.0
l.							0.0
							0.0
	l						0.0
		Subtotal		5.7	21,573	0	
Tho	se costs associated with progr	am management should be indicated by place	ment of an *.		F	Personnel Total	\$20.2
Trav	el Costs:		Ticket	Round	Total	Daily	Proposed
PM	Description		Price	Trips	Days	Per Diem	FFY 1996
							0.0
ļ	One trip to Anchorage for EV	OS Workshop	224	1	5	138	0.9
	One trip to Anchorage for EV	OS Workshop	224	1	3	138	0.6
		•					0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
	L						0.0
Tho	se costs associated with progr	am management should be indicated by place	ment of an *.			Travel Total	\$1.5
·			<u> </u>		1		
		Project Number: 98058 B				1	ORM 3B
1	1000	Project Title: Cutthroat Trout, Dolly V	arden Char H	Habitat Resto	ration,		Personnel
	1996	I ake Elsner Area					& Travel
1							
		Alency. Osca				L	DETAIL

#### 1996 EXXON VALDEZ TRUS .... COUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

Contractual Costs:		Propos	sed
Description		FFY 19	96
Report publicatio	n/printing	1	.5
When a non-trustee or	rganization is used, the form 4A is required. Contractual	Total \$1	.5
Commodities Costs:		Propos	sed
Description		FFY 19	96
	Commodities	Total \$0	).3
1996	Project Number: 9 <del>6059 B</del> Project Title: Cutthroat Trout, Dolly Varden Char Habitat Restoration, Lake Elsner Area Agency: USFS	FORM 3B Contractual & Commodities DETAIL	& s

## 1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

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New Equipment Purchases:		Number	Unit	Proposed
Description	· · · · · · · · · · · · · · · · · · ·	of Units	Price	FFY 1996
				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 accessisted with a	enlacement equipment should be indicated by placement of an P	Nour Fr	winment Tetel	0.0
Eviating Equipment Hasses	epiacement equipment should be mulcated by placement of an n.		Autor Alter	0.05
Existing Equipment Usage:			ivumber of Linita	inventory
Description				Allency
				U
-				
		1		l
	Project Number: 96058 - B		F	ORM 3B
1006	Project Title: Cutthroat Trout, Dolly Varden Char Habitat Resto	pration,		quinment
1990	l ake Fisner Area			
	Ayency. Usrs		L	<u> </u>

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96177B

# Cutthroat Trout, Dolly Varden Char Habitat Restoration, Port Fidalgo and Port Gravina Area

Project Number:	96177B
Restoration Category:	General Restoration
Proposer:	USFS, Cordova Ranger District
Lead Trustee Agency:	USFS
Duration:	3 years
Cost FY 1996:	31.6
Cost FY 1997:	Dependent on the results in FY 1996
Cost FY 1998:	Dependent on the results of previous years
Geographic Area:	Prince William Sound, Port Fidalgo and Port Gravina area, 20 miles northwest of Cordova.
Injured Resource:	Cutthroat trout, Dolly Varden char

## ABSTRACT

Timber harvests in the Port Fidalgo and Port Gravina area, 20 miles northwest of Cordova, may have affected cutthroat trout and Dolly Varden char habitat. The Cordova Ranger District proposes to work with the Tatitlek Corporation to survey the area and determine if there are any existing or potential impacts. If problems are identified, plans for restoration projects will be developed.

## INTRODUCTION

In recent years there has been increasing concern over the cutthroat trout and Dolly Varden char populations in the Prince William Sound area. Studies by the Alaska Department of Fish and Game show that these species were adversely affected by the *Exxon Valdez* oil spill (Hepler et al. 1993). Local fishermen have also reported decreased numbers of fish, which has led to sportfishing closures and reduced bag limits throughout the area. It seems that the best long-term method of preserving these stocks is to protect and restore the habitats these species utilize.

At the present time it is not known whether timber harvest in the Port Fidalgo and Port Gravina area has affected their habitat or not, however, it is important to identify and fix problems, such as erosion around

culverts or roads, before the situation gets worse. The Cordova Ranger District proposes to work with the Tatitlek Corporation to identify any areas where restoration projects may be needed. Such work could include rehabilitating stream crossing sites, closing and reseeding roads that are no longer needed, or improving the riparian vegetation. If it is found that there are restoration opportunities, plans for rehabilitation projects can be developed for FY 1997.

The role of the Forest Service would be to take the lead in contacting the Tatitlek Corporation and other concerned groups and developing a plan for examining the areas to see if problems exist. If restoration work is necessary, the Forest Service could help develop proposals, assist with the permit process, and work on the environmental analysis. Since the work would be on Tatitlek Corporation land, the corporation would be responsible for hiring crews and carrying out the project in FY 1997. The role of the Forest Service at this point would be to provide technical assistance if needed.

## **NEED FOR THE PROJECT**

## A. Statement of Problem

Studies by Hepler et al. (1993) showed not only that cutthroat trout had been adversely impacted by the *Exxon Valdez* oil spill, but also that many of the populations are quite small, making them more vulnerable to negative effects. The numbers of Dolly Varden char are generally higher, but their survival rates were found to be affected more by the oil spill than the cutthroat trout. Fishermen in the Cordova area have also reported that cutthroat trout populations have declined. It is not known to what degree either species has been affected or if they have made any recovery from the oil spill or other impacts. However, until more is known, it will be especially important to protect the stocks and habitat that remain and restore those areas which have been degraded.

Although these areas have been logged, it is not known if the fish habitat has been affected. It is possible that there are no problems. However, if there are areas where erosion from road crossings or other problems exist, it would be best to identify those problems now before they get worse. This project would ensure that we are aware of any potential threats to fish habitat and have a plan for restoring the degraded habitat.

## B. Rationale

Although this area was not directly affected by the *Excon Valdez* oil spill, Hepler et al. (1993) found that cutthroat trout and Dolly Varden char can stray considerable distances and move into different watersheds. Thus, it seems judicious to try to protect or restore habitat throughout the Sound. Protection of the habitat is also the best way to help maintain healthy populations and protect them against adverse impacts in the future, which is one of the objectives of the cutthroat trout/Dolly Varden char recovery plan.

## C. Summary of Major Hypotheses and Objectives

The main objectives for FY 1996 are to develop a memorandum of understanding with the Tatitlek

Corporation to conduct a joint survey of the logged areas around Port Fidalgo and Port Gravina, identify any problems that may affect fish habitat, and develop a restoration plan, if necessary. If the Tatitlek Corporation agrees with this project, restoration work would be carried out in FY 1997 by crews hired by the corporation. At this point it is thought that most of the work would involve removing culverts at stream crossings that are no longer needed, rehabilitating stream banks at those sites, and controlling any erosion that introduce sediment into the streams. Monitoring would occur in FY 1998.

## **D.** Completion Date

Restoration work should be completed in one field season (FY 1997), and the monitoring should be conducted in FY 1998.

## **COMMUNITY INVOLVEMENT**

The restoration work and hiring of the crews would be conducted by the Tatitlek Corporation. Generally, other Native corporations have expressed a hiring preference for corporation members, Native Alaskans, and local residents. Thus, it is expected that the local community would be directly involved in the project.

#### FY 96 BUDGET

Personnel	23.1
Travel	1.5
Contractual	1.5
Commodities	1.9
Equipment	0.0
Subtotal	<b>28</b> .0
Gen. Admin	3.6
Total	31.6

## **PROJECT DESIGN**

## A. Objectives

The main objective for FY 1996 is to conduct stream surveys and other watershed analyses to identify factors which could have adverse effects on cutthroat trout habitat. It is thought that the most likely problems would be bank erosion at stream crossings or other factors leading to stream sedimentation. If problems are identified, a restoration plan would be developed. In FY 1997 the restoration work would be carried out. In FY 1998 the work would be monitored.

## B. Methods

Streams in logged areas will be identified from recent aerial photographs. Crews will walk up each stream

and note the following:

- 1. Presence of suitable spawning or rearing habitat.
- 2. Bank erosion or other conditions which may lead to stream sedimentation.
- 3. Adequacy of riparian vegetation for input of large woody material, shading.
- 4. Presence of excessive slash or other logging debris.
- 5. Other unfavorable conditions.

Crews will also survey the roads to identify conditions which could affect streams located downhill from the roads. This could include:

- 1. Culverts or ditches which concentrate runoff and are causing erosion.
- 2. Destabilized slopes.
- 3. Areas without vegetation which could be eroded.
- 4. Roads which channel runoff and sediment to the streams.

If problems are identified, the location will be recorded and a brief restoration prescription will be written. Restoration measures could include:

- 1. Stabilizing or protecting eroding banks with rip-rap or log structures.
- 2. Revegetating bare or eroded areas with native plants.
- 3. Removing culverts and rehabilitating stream crossing sites.
- 4. Closing unneeded roads and revegetating.

It should be noted that buffer strips were left around most streams, so the work may be limited to stream crossing sites and other road work.

## C. Contracts and Other Agency Assistance

No contracts would be needed for the actions proposed in FY 1996. Depending on how the Tatitlek Corporation would like to handle the restoration work, contracts might be needed in FY 1997.

We would coordinate our efforts with the Alaska Department of Fish and Game, Habitat Division and incorporate any suggestions, advice, or ideas they may have. Otherwise, no other agency assistance is needed.

## D. Location

The project area would be in the Port Fidalgo and Port Gravina area, approximately 20 miles northwest of Cordova, Cordova C-7 quadrangle, T 13S, R 7W, sections 15-34 and R 8W, sections 13-34. The benefits of the project would be realized in a widespread area. Since this project will also protect coho, pink, and chum salmon, commercial and recreational fishers from Cordova and elsewhere will benefit. Other anadromous cutthroat trout or Dolly Varden char populations in Prince William Sound may benefit, depending on the amount of straying. The residents of Cordova would also benefit from the job opportunities created by this project, depending on how the hiring is conducted.

## SCHEDULE

## A. Measurable Project Tasks for FY 96

October to December 1995:	Meet with participants, develop MOU.
January to March 1996:	Hire study crew, develop data sheets, logistics.
May to July:	Conduct surveys, make initial prescriptions.
July	Revisit sites; finalize work plans.
August to September	Annual report.

## B. Project Milestones and Endpoints

- 1. Surveys and prescriptions will be completed by July 1996.
- 2. Restoration work will begin May 1997 and end in September 1997.
- 3. Monitoring will be completed by July 1998.

## C. Project Reports

Since this project does not include lengthy data analysis and the project work will occur in the summer months, annual reports can be completed by the end of each fiscal year.

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

At this point we do not foresee any opportunities for coordination or integration with other agencies or restoration efforts.

Since this project would be on land owned by the Tatitlek Corporation, it would not be part of the normal Forest Service management responsibility. The Cordova Ranger District is proposing this project to promote the overall health of the cutthroat trout and Dolly Varden char populations in the area and to help maintain angling opportunities on non-Forest Service land to keep fishing pressure dispersed.

#### ENVIRONMENTAL COMPLIANCE

The proposal for the actual restoration work would have to be reviewed by the State of Alaska Division of Governmental Coordination and the U.S. Army Corps of Engineers. The Corps permit for the project should be covered by the Nationwide Permit Program, permit numbers 13 and 27, which authorize erosion control work and restoration of riparian areas. The U.S. Forest Service could conduct the NEPA review, which would necessitate a biological evaluation, public scoping, and a categorical exclusion.

#### PERSONNEL

Project Leader:

Ken Hodges USDA Forest Service, Cordova Ranger District P.O. Box 280, Cordova, AK 99574 (907) 424-7661 (telephone) (907) 424-7214 (Fax)

Responsibilities: Plan surveys, train crews, analyze data, make recommendations for restoration work.

Ken Hodges is a fisheries biologist on the Cordova Ranger District. He has a B.S. degree in fisheries from Humboldt State University. Before coming to the District in 1989, he had worked as a seasonal employee for the Oregon Dept. of Fish and Wildlife and conducted a one-year study on steelhead genetics in Northern California. In Cordova he has worked as a fisheries technician and now as a biologist.

Project Manager:

Ray Thompson USFS Chugach National Forest 3301 C Street, Suite 300 Anchorage, AK 99503 phone (907) 271-2500

**Date Prepared** 

#### Literature Cited

Hepler, K.R., P.A. Hansen, and D.R. Bernard. 1993. Impact of oil spilled from the *Excon Valdez* on survival and growth of Dolly Varden and cutthroat trout in Prince William Sound. Alaska Department of Fish and Game. Anchorage, AK.

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

	Authorized	Proposed						
Budget Category:	FFY 1995	FFY 1996						
Personnel		\$23.1						
Travel		\$1.5						
Contractual		\$1.5						
Commodities		\$1.9		ta <u>lin Arr</u> i,	al an an the second	under det in de la companya de la co		
Equipment		\$0.0		LONG F	RANGE FUNDIN	IG REQUIREMEN	NTS	
Subtotal		\$28.0	Estimated	Estimated	Estimated	Estimated	Estimated	Estimated
General Administration		\$3.6	FFY 1997	FFY 1998	FFY 1999	FFY 2000	FFY 2001	FFY 2002
Project Total		\$31.6	\$0.0	\$0.0	\$0.0	\$0.0	\$0.0	\$0.0
Full-time Equivalents (FTE)		0.5						
			Dollar amount	s are shown in	thousands of c	dollars.		
Other Resources			•					
	,							
	Project Num	ber: 90043	96177	В		[		FORM 3A
	Project Title	Cutthroat T	rout . Dolly V	arden Char I	Habitat Resto	pration. Port		
1996	Eidolao and	Port Grouino	Aroa					
	Fidaigo and	FUL GIAVINA	MIEd					PROJECI
	Agency: U	SFS						DETAIL
Prepared: 14	५२ã/92 [∟]							F/1/95

# 1996 EXXON VALDEZ TRUSTER JOUNCIL PROJECT BUDGET

October 1	1,	1995 -	September	30,	1996
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Pers	onnel Costs:		GS/Range/	Months	Monthly		Proposed
PM	Name	Position Description	Step	Budgeted	Costs	Overtime	FFY 1996
	D.Schmid	Project Leader	GS-11	0.7	4,620	0	3.2
	J. Hodges	Project Support	GS-9	1.0	3,864	0	3.9
	GS-7 -one	Fisheries tech	GS-7	1.4	3,339	0	4.7
	GS-5-one	Fisheries tech	GS-5	1.4	2,058	0	2.9
	GS-4-one	Fisheries tech	GS-4	1.4	1,764	0	2.5
*	R. Thompson	Program manager	GS-13	1.0	5,928		5.9
							0.0
							0.0
	•						0.0
							0.0
1							0.0
ļ	L						0.0
<b> </b>		Subtotal		. 6.9	21,573	0	
Tho	se costs associated with progr	am management should be indicated by place	ment of an *.		F	Personnel Total	\$23.1
Trav	el Costs:	·	Ticket	Round	Total	Daily	Proposed
PM	Description		Price	Trips	Days	Per Diem	FFY 1996
					_		0.0
A	One trip to Anchorage for EV	OS Workshop	224	1	5	138	0.9
	One trip to Anchorage for EV	OS Workshop	224	1	3	138	0.6
							0.0
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1							0.0
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							0.0
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							0.0
							0.0
				ll		Travel Tetal	0.0
Ino	se costs associated with progra	am management should be indicated by place	ment or an ".			ITAVEI IOLAI	C.1¢
						r	
	· · ·	Project Number: <del>96043 D</del>					FORM 3B
	1006	Project Title: Cutthroat Trout , Dolly \	Varden Char	Habitat Resto	pration,		Personnel
	1990	Port Fidalgo and Gravina Area				1	& Travel
1		Agency: USFS					DETAIL
L						L	

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

Contractual Costs:			Proposed
Description			FFY 1996
Report publication/pri	inting		1.5
When a non-trustee organi	ization is used, the form 4A is required. Co	ntractual Total	\$1.5
Commodities Costs:			Proposed
Description			FFY 1996
Food for 105 days @	) \$15.00 a day		1.6
	Com	modities Total	\$1.9
1996	Project Number: <del>96049 D</del> Project Title: Cutthroat Trout , Dolly Varden Char Habitat Restoration, Port Fidalgo and Port Gravina Area Agency: USFS	F Cor Co	ORM 3B htractual & mmodities DETAIL

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

New Equipment Purchases:		Number	Unit	Proposed
Description		of Units	Price	FFY 1996
				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 r	eplacement equipment should be indicated by placement of an R.	New Ec	uipment Total	\$0.0
Existing Equipment Usage:			Number	Inventory
Description			of Units	Agency
				0
	۱. King and a second			
	f		<u></u>	
	Project Number: 9 <del>6943 &gt;D</del>		F	ORM 3B
1006	Project Title: Cutthroat Trout, Dolly Varden Char Habitat Resto	oration. Port		
1990	Fidaloo and Port Gravina Area	,		OETAN
				DETAIL
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# Second Growth Forest Habitat Enhancement for Injured Wildlife Species

Project Number: 96178			
Restoration Category:	General Restoration		
Proposer:	USFS		
Lead Trustee Agency:	USFS		
Duration:	5 yrs		
Cost FY 96:	\$84,300		
Cost FY 97:	\$90,500		
Cost FY 98:	\$60,000		
Cost FY 99:	\$60,000		
Cost FY 00:	\$60,000		
Geographic Area:	Prince William Sound		
Injured Resources/Service:	Bald Eagle, River Otter, Marbled Murrelet, Harlequin Duck		

## ABSTRACT

The Prince William Sound area has several watersheds on National Forest System lands where timber harvest occurred in the early 1970's. These were done without an understanding of optimum stand structure for wildlife populations. This project has the potential to improve habitat for river otter, marbled murrelet, harlequin duck and bald eagle by accelerating succession and developing forest stand structure beneficial to wildlife species faster than natural forest succession. Habitat for old-growth dependent species such as river otter, marbled murrelet, harlequin duck, and bald eagle, whose populations were proven to be damaged by the 1989 Oil Spill, can be improved with this project.

## INTRODUCTION

One possible way to counteract the effects of the 1989 oil spill is to restore habitat for injured species on land. Prince William Sound old-growth forests are characterized by the presence of large diameter trees, multilayered canopies, a range of tree diameters, and understory vegetation. These stands are generally well past the age of maturity as evidenced by dead and dying trees, snags and other downed woody material. These characteristics are important to several of the injured species during all or a portion of their life cycle.

Approximately 2,525 acres of second growth in Prince William Sound are on National Forest System lands. Approximately 1,733 acres have been harvested on Montague Island, 233 acres have been harvested on Hinchinbrook Island, and 122 acres were clearcut harvested in Puget Bay. Stands harvested range from pure Sitka spruce to stands composed of 40% Sitka Spruce and 60% hemlock. The majority of these harvested areas are located on lower slopes from 0 to 100 feet in elevation. Two of the areas on Montague Island extend to 700 feet in elevation. The harvested areas range in size from 4 acres to 201 acres with a mean size of 42 acres. Most of these areas are located along bays and streams which could provide habitat for the injured species if the necessary old growth components were available.

Forest successional patterns after timber harvest vary with soil type and degree of soil disturbance. Sites revegetate rapidly. Conifer regeneration prevails where soil disturbance is slight to moderate, favoring western hemlock or Sitka spruce depending on severity. An alder stage is most likely to occur in areas of severe soil disturbance and in alluvial bottom-land sites. Residual shrubs respond dramatically. Their growth increases exponentially, with a peak in cover and biomass about 15 to 25 years after the overstory is removed. Conifers begin to overtop shrubs by years 8 to 10 years. By year 15, crown closure begins, and it is complete by about year 25. Subsequent decline in vascular understory is rapid and elimination occurs in 25 to 35 years. This condition is projected to last to stand age 140 to 160 years and is attributed to subdued or low level light conditions caused by the forest canopy structure. Absence of an understory during the major part of the rotation is likely to be the single most important concern from a wildlife habitat standpoint if a second-growth forest is not managed.

## NEED FOR THE PROJECT

#### A. Statement of Problem

Two of the most pressing problems of second growth management and wildlife are snow interception and understory development. These are a direct consequence of tree density and canopy closure in young stands. Without management inputs, second growth stands will require 250 years to begin acquiring old-growth characteristics. Managing second growth in Alaska's coastal rain forest with emphasis for fish and wildlife has been a documented practice since the early 1980's in Southeast Alaska. Techniques used to advance succession and maintain the understory component in Southeast Alaska have not been applied to second growth forests in Prince William Sound.

Non-commercial thinning is the major treatment available for altering these stand structure characteristics. Approximately 2,525 acres of second growth resulting from timber harvest in Prince William Sound is available for treatment. It is necessary to determine which acres should be treated to most benefit the injured species. During the past 20 years extensive research and trials have been made throughout Southeast Alaska to determine if the wildlife habitat value of second growth can be enhanced through silviculture techniques such as thinning, gap formation or pruning. Although much of this research may directly apply to Prince William Sound, it is essential that a careful assessment be conducted first before large commitments to management are made due to the potentially high costs

and uncertain results. Because of the climatic, soils, and overall ecological conditions in the Sound relative to Southeast Alaska, it is possible that treatment response might be slower or perhaps fundamentally different. A survey of successional development and thresholds of canopy openings for the maintenance of understory diversity in these forests is needed to assess the potential for silvicultural improvement in habitat structure in the region.

## B. Rationale

This project has the potential to improve habitat for harlequin duck, marbled murrelet, river otter and bald eagle by exploring ways to promote successional trends and provide components of old-growth stands earlier than 250 years from harvest. These wildlife species were proven to be damaged by the 1989 Oil Spill. This project can rehabilitate habitats for those injured species.

The results from the project will augment other thinning and stand development studies. the project will test how general and applicable the previous studies are to other regions and specifically to lower sites and a more extreme climatic condition.

## C. Summary of Major Hypotheses and Objectives

Sitka spruce and western hemlock migrated to Prince William Sound about 3,000 years ago. The coastal forest in Prince William Sound is reduced in stature, productivity and species richness compared to Southeast Alaska. Forests in Prince William Sound have lower site indices than predicted from the latitude relationships of Harr and Harris. This may be attributed to the heavy snow packs, younger, shallower soils and relatively cool, short growing season. The heavy snow packs persist into the growing period, effectively cooling the soils, reducing the biological activity, and slowing soil development. This difference may cause unexpected results during stand manipulation. In most cases and as experienced in Southeast Alaska, thinning by delaying crown closure should prolong existence of understory vegetation on cutover areas.

Prescriptions for timber production and those for wildlife habitat improvement may be different depending on the desired stand structure. Thinning for wildlife objectives might maintain lower stocking levels of trees to encourage development and longevity of the shrub understory layer. Heavy thinning may release conifer regeneration which would defeat the purpose of the thinning from a wildlife point of view. Wildlife thinning should be conducted early enough to maintain a vigorous understory layer. In contrast, thinning to meet timber production goals would be delayed until crown closure so competing plants and trees are shaded out.

The purpose of this project is to provide mitigation through habitat enhancement for identified injured wildlife species in Prince William Sound. The objectives are:

- 1. Maintain understory vegetation components throughout the successional stages of second growth and manage for characteristics important to injured species.
- 2. Increase successional trends in key wildlife habitat areas to develop old growth structure. Approximately 2,500 acres of second growth habitat will be evaluated for enhancement opportunities.

3. Establish plots on a subset of sites to determine how quickly vegetation responds to treatment and to determine the overall successional pattern on productive sites.

## D. Completion Date

This project will be completed in fiscal year 2000.

## COMMUNITY INVOLVEMENT

It is anticipated that as potential areas are identified for treatment, the public will be asked to participate in the identification of issues and concerns. Currently the Forestry Sciences Lab in Juneau has been involved in project identification and development.

#### FY 96 BUDGET

Personnel	49.9
Travel	11.0
Contractual	13.0
Commodities	2.0
Equipment	0.0
Subtotal	75.9
Gen. Admin.	8.4
Total	84.3

Travel costs consist of small aircraft charters, WCF boat rentals and field per diem besides 2 trips to Anchorage for principle investigator. Contractual is a contract for low aerial photography. In kind services consist of salary of participants from the Forestry Science Lab in Juneau and preliminary work completed by the District to date.

## **PROJECT DESIGN**

#### A. Objectives

Approximately 2,500 acres of second growth habitat will be evaluated for enhancement opportunities. Successional trends will be increased in key wildlife habitat areas to develop old growth structure. Following is a description of the injured species habitat requirements that this project will address. Site prescriptions will be made to enhance these habitat characteristics.

#### **River Otter (Lutra canadensis)**

Areas of riparian vegetation provide river otter resting and denning places as well as protective cover for traveling. Natural cavities in old-growth forests, rock cavities, and burrows or lodges of other animals are used for dens. Latrine sites are located along shorelines in old-growth forest areas and adjacent to suitable feeding areas. Old-growth forested areas are also used as resting places as otters travel along their home ranges. Home ranges vary with quality of habitat and can vary from 7 to 40 kilometers.

#### Harlequin Duck (Histrionicus histrionicus)

In May, paired harlequins congregate at mouths of anadramous fish stream and fly inland to search for nest sites. Typically nests are located along shallow rivers and streams with gravel or rocky substrates. Nest sites are located under dense vegetation on steep banks in mature forests. Harlequins may return to the same nest site in consecutive years. Slow stretches on lee sides of bends in streams are used by broods for feeding and resting.

#### Marbled Murrelet (Brachyramphus marmoratus)

Current data suggests that most marbled murrelets nest in mature forests. Most have been located in large conifers, but ground nests have been recorded. They are solitary nesters and have been located as far as 40 to 50 km from the coast.

#### Bald Eagle (Haliaeetus leucephalus)

Bald Eagle nests are usually located in older, larger trees. Coastal areas with more than one nest per mile are considered to be good nesting areas. Nests are usually used for more than one season. Bald eagles prefer large diameter mature trees for nest and perch sites. Typical nest trees are at least 400 to 500 years old.

By accelerating the succession of harvested stands to obtain old-growth characteristics sooner than if no treatment was done may help offset the impacts of the spill in the long term. Following is the characteristics of the old-growth forest types which will be the desired future condition of the potentially treated stands.

#### Sitka Spruce - Alluvial Forest Type.

This forest type is most often associated with riparian ecosystems. Sitka spruce dominates the stand with western hemlock, mountain hemlock and Sitka alder occurring as minor components. Stands are relatively open grown with tree canopy cover ranging from 30 to 60%. Stands are typically multilayered, with mid canopy layer trees growing in the open. The height of the upper canopy often ranges from 90 to 110 feet and large diameters range from 16 to 40 inches. Trees have been recorded exceeding 160 feet in height and diameters up to 63 inches on the best sites. Natural regeneration of spruce is sporadic and usually occurs only on microsites. Down material as "nurse logs" is important to conifer regeneration. Tall shrub cover (salmonberry, blueberry, and devil's club) dominates the ground vegetation, often exceeding 60% cover. Forb cover (five leaf bramble, bunchberry, and skunk cabbage) is moderate. Soils are deep, generally well drained, with thin organic layers. Soil disturbance through flooding is frequent and restricts soil profile development. The thin organic layers are easily disturbed which favors re-establishment of salmonberry, devil's club, and alder. This forest type is usually associated with riparian habitats where it provides bank stability, cover and woody material input into streams. Devils club, currant, and salmonberry fruit production is relatively high. Salmon, skunk cabbage, and berry production cause this type to be heavily used by

brown bear. Snags, though few in number, tend to be relatively large and provide good habitat for cavity dwellers.

## Sitka Spruce - Other Forest Type.

The Sitka Spruce - Other Old Growth Type is most often found in the hemlock spruce coastal zone. Many stands occur in a narrow band along the coast. Sitka spruce is the major tree component, however western hemlock and mountain hemlock are components of the overstory. Tree canopy cover ranges from 70 to 80%, is typically multilayered, and contains large branched, deep crowned hemlock and spruce. Western and mountain hemlock are dominant understory trees. The height of the upper canopy ranges from 70 to 110 feet and large trees diameters often range from 13 to 30 inches. Shrub cover (blueberry, salmonberry, rusty menziesii and devils club) is moderate (40 to 50%) and forb cover (five leaf bramble, bunchberry, foam flower, and skunk cabbage) often exceeds 30%.

## Western Hemlock - Well Drained Forest Type.

The Western Hemlock - Well Drained Old Growth Forest Type is common to the coastal areas of southcentral Alaska. Most stands occur on well drained shallow soils formed by either colluvial or alluvial action and are located at elevations below 600 feet. Western hemlock dominates the overstory in this type but stands are rarely pure western hemlock. Regeneration consists primarily of western hemlock with sparse amounts of Sitka spruce and mountain hemlock. Tree canopy cover often ranges from 70 to 80%, is multilayered, and contains relatively large branched, deep crowned hemlock. The height of the upper canopy often ranges from 70 to 120 feet and large tree diameters range from 14 to 35 inches. Shrub cover is usually moderate (40-50%) and is dominated by blueberry. However, it also contains salmonberry, rusty menzesii, and devils club. Forb cover (five leaf bramble, bunchberry, wintergreen and skunk cabbage) often exceeds 30%. On devil's club sites, herbaceous vegetation is abundant. Common ferns are lady, deer, and shield fern. Downed woody material is large and abundant, originating from both windthrow and tree breakage. Under natural conditions, stand closure occurs between age 30 and 40 years. Under these dense, closed canopy conditions, moss can dominate the understory through age 100 years. By age 150 years, the canopy has generally thinned to allow the understory to develop. If mineral soil is exposed, Sitka alder is likely to dominate the site for a period of time. Large spruce and hemlock trees effectively intercept snow. Understory vegetation is relatively abundant and generally available for forage during winter. Large standing trees provide good nest and forage sites for cavity dwellers. Large down woody material and elevated root systems are often present and provide good forage, resting, and denning sites for a variety of wildlife.

## **Mountain Hemlock - Low Elevation Forest Type**

The Mountain Hemlock - Low Elevation Old Growth Forest Type is often found on poorly drained soils with deep organic layers and often occupying coastal headwaters, lowland rolling hills, and muskegs. It occurs from sealevel to 500 feet elevation in Prince William Sound. Mountain hemlock is the dominant overstory tree. Western hemlock is the most consistent associated conifer and Sitka spruce is a minor component along beach fringes. The height of the upper canopy often ranges from 50 to 70 feet and large tree diameter may range from 7 to 21 inches. Large trees as tall as 110 feet and 40 inches diameter have been recorded on better sites. A dense tall shrub layer of blueberry

along with rusty menziesii, salmonberry, and devil's club often exists with a low shrub layer of crowberry and bog blueberry. The forb layer often includes skunk cabbage, five-leaf bramble and twisted stalk. Snags are highly variable both in size and number, and are considered short lived due to the species characteristics, high snow loads, wind, and avalanches. Tree ages range from 400 to 600 years with a Krumholz appearance. Mountain hemlock forests are important habitats for deer and brown bear during the summer months.

## B. Methods

This project will be accomplished by the following sequence of events:

- 1. Inventory existing data base. This consists of compiling existing data on Prince William Sound second growth, establishing date of harvest, and entering information into GIS data base. It also consists of compiling existing information on injured wildlife habitat needs and entering appropriate information into the GIS data base.
- 2. Inventory existing habitat. This will be accomplished by low level aerial photography of all second growth sites and field sampling to determine existing vegetation community type and site potential. Field data collected will include use of data collection cards designed to sample old-growth to determine how much "old-growth" characteristic are still present in the stands. Plant association information, stem densities, species distributions, and percent ground cover by species will also be collected. Occurrence of insect and diseases will also be recorded to identify potential problems with prescribed treatments. Approximately one week will be spent surveying clearcuts from 14 to 42 years old in which vegetation structure and canopy architecture will be surveyed and measured. Radiometers and other instrumentation will be used to determine how dense a canopy understory plants can grow under and how quickly vegetation responds to disturbance. Five or more sites will be examined along this canopy density and stand age gradient to determine the overall successional pattern on productive sites. Small plots will be established on a subset of sites to determine understory cover, biomass, and growth as related to stand age, density, size, and percent light penetration through the over story canopy.
- 3. Define vegetation objectives. After the second growth areas have been described, vegetation objectives will be established specific to the targeted species.
- 4. Prepare site prescriptions. Prescriptions will be developed identifying recommended treatment to meet the vegetation objectives. Treatment options could consist of pre-commercial thinning at varied spacing to maintain understory vegetation throughout the rotation. Coordination with people in Southeast Alaska working on similar projects will be maintained to gain insight for potential treatments.
- 5. Conduct environmental analysis. An environmental analysis will be conducted prior to a decision as required by the National Environmental Policy Act.
- 6. Implement prescriptions and establish study plots for monitoring. If the decision is made to implement the recommended treatment after the environmental analysis this project could be

accomplished over the following three years.

## C. Contracts and Other Agency Assistance

The Forest Sciences Lab in Juneau will be providing assistance in designing thinning regimes and installing permanent monitoring plots.

## D. Location

The project will take place in Prince William Sound. Final location of the project will be determined during feasibility study. Potential locations include the northeast side facing side of Montague Island or Hinchinbrook Island.

## SCHEDULE

## A. Measurable Project Task for FY 96

January 1 - February 15:	Inventory existing database
June 1 - 15:	Develop low aerial photography
June 1 - September 1:	Inventory habitat
September 1 - 15:	Define vegetation objectives
September 15 - 30:	Write site prescriptions
FY 97 and beyond	

November 1 - March 1:	Conduct environmental analysis and documentation
March 1 - May 15:	Prepare contracts and obtain bids
summer 1997-2000:	Implement prescriptions by contract

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

February 15, 1996:	Complete inventory of existing data and map existing data	using GIS
May 31, 1996:	Data needs determined and sampling design developed	ŧ
September 1, 1996:	Prescription options developed by coordinating with FSL. and data collection, analysis and summaries completed.	Field work
September 15, 1996:	Desired Future conditions defined, objectives developed.	
September 30, 1996:	Prescriptions completed	
March 3, 1997:	Environmental analysis complete	
1997:	Contract prepared and awarded to implement prescription	

## C. Project Reports

Forestry Sciences Lab personnel will submit a summary report to the Cordova Ranger District on observations and implications to second growth management in the Fall of 1996. Preliminary data will be pooled with the data from Southeast Alaska to compare understory characteristics in relation microclimate.

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

The preliminary results of this project will be incorporated by the Forestry Sciences Lab in Juneau with existing studies conducted in Southeast Alaska. Results can be compared to determine how general and applicable the results are to other regions and specifically to lower sites and a more extreme climatic condition.

#### ENVIRONMENTAL COMPLIANCE

An environmental analysis and documentation will be completed before any vegetation manipulation takes place. Analysis is anticipated to take place in FY 97.

#### PERSONNEL

Daniel W. Logan, Wildlife Biologist, Cordova Ranger District, has been employed with the Forest Service for 17 years. He has held positions in Sitka, Petersburg, Seward and Cordova as fisheries biologist, wildlife biologist and District Wildlife Staff Officer. Mr. Logan has Bachelor Degrees in Wildlife and Fisheries from Humboldt State University.

Susan E. Kesti is a certified silviculturist for the Chugach National Forest. She has worked for the Forest Service for 18 years in Oregon, Sitka, Seward, and Cordova. Before working for the Forest Service, Ms. Kesti was a research assistant at the University of Idaho working on the Intensive Timber Culture Project. She has held positions as a Forester, Environmental coordinator, and District Timber Staff. She has a Bachelors Degree in Forestry from Michigan Technological University and has completed graduate level course work at the University of Idaho, University of Oregon, and the University of Washington.

Project Leader:

Daniel W. Logan USFS, Cordova Ranger District, Chugach National Forest P.O. Box 280 Cordova, AK 99574 (907)424-7661 Fax (907) 424-7214

Project Manager:

Ray Thompson USFS Chugach National Forest 3301 C Street, Suite 300 Anchorage, AK 99503 phone (907) 271-2500 FAX (907) -271-3992

Date prepared

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#### **1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET**

October 1, 1995 - September 30, 1996

	Authorized	Proposed						
Budget Category:	FFY 1995	FFY 1996						
Personnel		\$49.9						
Travel		\$11.0						
Contractual		\$13.0						
Commodities		\$2.0						
Equipment		\$0.0		LONG F	RANGE FUNDIN	G REQUIREME	NTS	
Subtotal	\$0.0	\$75.9	Estimated	Estimated	Estimated	Estimated	Estimated	Estimated
General Administration		\$8.4	FFY 1997	FFY 1998	FFY 1999	FFY 2000	FFY 2001	FFY 2002
Project Total	\$0.0	\$84.3	\$90.5	\$60.0	\$60.0	\$60.0	\$60.0	
Full-time Equivalents (FTE)		1.0		사람 문제가 가지 가지 않는다. 바라 가지 하는 것 같은 것 같은 것	n in twenty in Alternation			
			Dollar amount	s are shown in	thousands of c	Iollars.		
Other Resources			· ·		•			
[]	Project Num	ber:					[	FORM 3A
	Project Title:	: Second Gro	owth Forest F	labitat Enhai	ncement For			AGENCY
1996	Injured Wild	life Species						PROJECT
	Agency: US	SFS						DETAU
							L	DETAIL
Prepared: 1 of	4 L					]		/1/95

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1996	FXXON	VAI DEZ	TRUS'	CUNCIL	PROJECT	BUDGET
					11100-01	

Pers	onnel Costs:		GS/Range/	Months	Monthly		Proposed
PM	Name	Position Description	Step	Budgeted	Costs	Overtime	FFY 1996
	Sue Kesti	silviculturist	GS-11/6	5.0	4,400		22.0
	Dan Logan	Wildlife Biologist	Gs-11/6	2.0	4,400		8.8
	Don Youkey	Wildlife Biologist	GS-7/2	2.0	3,300		6.6
	Wildlife Technician	Wildlife Technician	GS-7/2	2.0	3,300		6.6
<b>*</b> .	R. Thompson	Program Manager	GS13	1.0	5,928		5.9
					1		0.0
							0.0
l							0.0
\$1							0.0
					1		0.0
ĺ.			1				0.0
							0.0
			Subtotal	12.0	21,328	. 0	
Tho	se costs associated with	program management should be indicat	ed by placement of an *.		<u> </u>	ersonnel Total	\$49.9
Trav	el Costs:		Ticket	Round	Total	Daily	Proposed
PM	Description		Price	Trips	Days	Per Diem	FFY 1996
	Air Charter to field sites	i	400	5	40	60	4.4
	WCF Boat Rental		3000	. 1	40	60	5.4
	flights to anchorage		200	2	6	135	1.2
							0.0
			1 1	1			0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
		·					0.0
Tho	se costs associated with	program management should be indicat	ed by placement of an *.			Travel Total	\$11.0
		Project Number:			1		FORM 3B
1		Project Title: Second Ground	th Forget Habitat Enhan	coment For		1	Personnel
ł	1996	roject nite. Second Glow					9. Transl

#### October 1, 1995 - September 30, 1996

Injured Wildlife Species

Agency: USFS

## 1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

Contractual Costs:		Proposed
Description		FFY 1996
Aerial Photo Contract Report publication/printing		10.0 3.0
When a non-trustee organization is used, the form 4A is required.	al Total	\$13.0
Commodities Costs:	T	Proposed
Description		FFY 996
Field supplies		2.0
Commoditie	s Total	2.0

#### 1996 EXXON VALDEZ TRUST___DUNCIL PROJECT BUDGET October 1, 1995 - September 30, 1996

New Equip	ment Purchases:	Number	Unit	Proposed
Descriptio	n	of Units	Price	FFY 1996
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
Those pur	chases associated with replacement equipment should be indicated by placement of an R.	New E	quipment Total	\$0.0
Existing E	quipment Usage:		Number	Inventory
Descriptio	n .		of Units	Agency
<u> </u>				
	Project Number:			ORM 3B
10	Project Title: Second Growth Forest Habitat Enhancement For			auinment
193	Injured Wildlife Species			DETAIL
	Agency: LISES			DETAIL
L			L	

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696179

trout.

# Relationships between in stream habitat attribute features and the USFS Alaska Region stream classification system channel types within Prince William Sound.

Project Number: Restoration Category:	74179 Research
Proposer:	USDA Forest Service
Lead Trustee Agency:	USFS
Cooperating Agencies:	
Duration:	2 years
Cost FY 96:	\$218,100
Cost FY 97:	\$40,300
Study Area:	Prince William Sound
Injured Resource/Service:	Pink salmon, cutthroat trout, Dolly Varden

## ABSTRACT

Channel types represent similar hydrological and geological similar or reaches of stream, they should also be relatively good descriptions of what is present for in stream fish habitat. Channel type interpretations should provide a quantitatively replicable measure for presence of instream spawning and rearing habitat. This project will further the understanding of the anadromous salmonid capability habitat relationships of the watersheds within PWS.

## INTRODUCTION

Pacific salmon and trout of Prince William Sound (PWS) have complex life histories that depend on both marine and freshwater environments. Rates of salmon mortality are highest during their early stream dwelling life forms. Because the mortality is often related to the condition and availability of the in-stream habitat, it is critical that habitats limiting to juvenile salmon are protected or restored. Basin wide in stream habitat surveys are essential to predict those habitat conditions that limit survival. In remote areas of Alaska it is impractical to physically survey all streams within a geographic area, therefore, a hierarchical approach lends itself to remote sensing techniques. This hierarchical approach, aided by photo interpretation, significantly increases the efficiency of large scale habitat inventories.

Preliminary data, collected on the Kenai Peninsula in 1991, suggests that channel types, as defined from aerial photographs, may be a predictor of at least several micro habitats found in a stream

channel. A multi-variate analysis of variance, indicated that channel types were a significant predictor (p < 0.0001) for eight of thirteen micro habitats (e.g. rapids, plunge pools, and dammed pools) as modified from Bisson (1981). Further data collection, for the EVOS Trustee Council Channel Type Classification Study (90505, Part C) indicated that channel types are a significant predictor of the amount of spawning and rearing habitat in a given segment of stream.

The existing data bases have been analyzed to firmly establish the relationship between aerial photo channel type interpretations, and the presence of in-stream spawning and rearing habitat. These analysis and report will serve as a basis for larger scale in-stream habitat surveys for oil spill affected areas in PWS. This project will further the understanding of the anadromous salmonid capability habitat relationships of the watersheds within PWS.

## NEED FOR THE PROJECT

## A. Statement of Problem

Anadromous fish were injured by oil spill contamination from the Exxon Valdez oil spill of March 1989. The oil spill affected anadromous fish in several ways; pink salmon had high egg and fry mortalities, reduced growth rates, and possible morphological abnormalities; and sockeye salmon suffered poor smolt survival due to over-escapement. Salmon also have exacting freshwater habitat requirements and are vulnerable to human-caused changes in habitat quality. The fish have adapted to the variable conditions in stream environments, and have segregated into stocks that are specifically adapted to local landscapes and possibly to local watersheds. The quantity of fish habitat in any stream depends on the condition of the area that it drains. Thus, watersheds are the basic unit of forested landscapes, they control to a large extent the populations of anadromous fish present and numbers of fish. Prince William Sound contains many islands with small watershed and short stream systems that together are believed to contain numerous stocks of salmon and anadromous trout and char. The populations of individual stocks in these short stream systems are small making them highly vulnerable to watershed disturbances that affect the quality of freshwater habitats. Significant disturbance of habitat in these watersheds, coupled with poor marine conditions and continued intense fish harvest could place these small populations at risk. Frissel, et al, 1986, advocated that classification of streams and the habitat contained would be useful in determining the impacts of land use practices, assessing basin wide cumulative affects of management practices on stream habitats, and provide useful generalized information on stream habitats from site specific data.

Inventories of habitat are a primary source of information for the evaluation of watershed conditions and the management and protection of aquatic resources. Data collected in comprehensive surveys can be used for the basis of habitat restoration and improvement programs and can be used to monitor changes in the quality and quantity of resources. The proposed study would provide the relationship between channel type designations and the presence of in-stream habitat features, e.g. various types of pools, , useful for both anadromous salmonid rearing and spawning. These data would provide valuable information in comparing relative values of all watersheds with PWS area, particularly those contained within comprehensive evaluation parcels.
## B. Rationale

Several survey methodologies have been developed to quantify and qualify habitat in section of streams (Bisson et al. 1982, Barber et al 1981, Hankin and Reeves, 1988). While these habitats surveys present a good estimation of what is present within that segment of stream, owing to habitat variability, the information is not a good predictor of overall habitat capability. Aquatic habitat inventory information on both habitat and fish provides important data for habitat protection and watershed analysis. If inventory procedures are quantitatively replicable, they become useful tools for monitoring trends in habitat condition over time.

Natural and human disturbances have additive effects on freshwater habitat, so analysis of effects and planning for activities that cause disturbance should be at a large spatial scale. Many management plans and protective measures for fish habitat on public and private land, however, are still applied on a smaller project area basis. Although project-level analyses are important, more attention should be given to larger spatial and temporal analyses so as to improve cumulative effects analyses. Watershed-scale planning, analysis, and implement of management (such as logging) can better pace fish habitat protection in the proper context. Focus of the watersheds studied will be on large and small parcels proposed for acquisition. A GIS based analysis will provide a tool for efficient comparison of the numerous watersheds draining in Prince William Sound. The study will quantify the natural range of variability for in-stream habitat features for watersheds within PWS. These natural ranges in variability for stream channel habitat features can provide important quantitative and measurable objectives for anadromous fish and resident trout habitats, which in turn can provide information needed in determining changes in anadromous fish habitat capability.

## C. Summary of Major Hypothesis and Objectives

We hypothesize that because channel types represent similar hydrological and geological similar or reaches of stream, they should also be relatively good descriptions of what is present for in stream fish habitat. If the channel types are good indicators of presence of in- stream habitat then we can establish natural ranges of variability for in-stream habitat features for channel types within PWS. We also hypothesize that if channel types are a good indicator of juvenile salmonid habitat, then they also can be used to predict relative rearing densities within in-stream habitats. If this is true then channel type differences are a means of estimating relative standing crops of juvenile salmonids.

## **D.** Completion Date

Final report will be completed by the end of FY 1997.

## COMMUNITY INVOLVEMENT

The villages of Chenega and Tatitlek will be visited to discuss the objectives of the study, and to

discuss their traditional knowledge( if the village elders are willing) of the watershed proposed for study.

## FY 96 BUDGET

Personnel	112.9
Travel	1.4
Contractual	77.5
Commodities	.5
Equipment	2.2
Subtotal	189.8
General Administration	22.4
Total	218.1

## **PROJECT DESIGN**

## A. Objectives

The purpose of the study is to characterize preselected habitat characteristics in an entire representative reach and /or watershed. The study will provide data from all habitat types and locations within a channel type or watershed. The study will identify all habitats and separate them into three levels; macro units, meso units, and micro units (Table 1). In addition to this application, visual observation techniques will be used to estimate juvenile salmonid densities in study streams using methods developed by Hankin and Reeves (1988). Finally, juvenile salmonid densities within habitat units and estimates total salmonid population by species will be evaluated.

## B. Methods

Field sampling will focus on stream reaches within watersheds identified as having significant spawning and rearing habitat value. These areas were identified in the Stream Channel Type study (in draft). All of these sites will be located within the oil spill area within Prince William Sound and be stratified by ecological subsection (Davidson, 1994) and ecological setting determined by mean geology, precipitation, and temperature.

A minimum of 10 study watersheds will be located. Based on interpretations from 1:15000 scale color aerial photos, and 1:63,360 scale USGS topographic quadrangle map, all streams within the study watersheds have been previously channel typed (Paustian, 1992) and digitized into a Geographical Information System. Channel typed streams will be ground verified. Sample sites will be located by demarcating the upper and lower limits of the stream channel type segments on 1:63,360 scale USGS topographic maps, and on the 1:15,000 scale color aerial photographs. Orienteering methods will be used to locate the downstream limit of the channel type break for each sample. For each channel type the field crews will sample habitat in two steps. The first will require sampling teams to classify individual habitat units by channel type and document visual observatior

habitat characteristics, such as water surface area and stream bottom composition. In the second step, the visual observations will be compared to actual measurement of habitat characteristics at a predetermined number of units. These measurements will be used to develop calibration ratios for the visual observations. These data will be entered into the Chugach National Forest database. Comparative analysis of the comprehensive evaluation parcels will be accomplished using models developed from habitat relations of the channel types.

Survey methods for streams will be adapted from Hankin and Reeves (1988) as per Bryant et al (1992). Each habitat survey will identify the length of stream surveyed for each channel type. Methods for estimating fish densities within PWS will follow methodologies established by Hankin and Reeves (1988). Methods for estimating total pool areas will be identified by methods developed by Bryant et al (1992). Here habitat type definitions are arrayed int three levels of detail (macro, meso, and micro levels). For example, all pools at any level must qualify as macro pools. Once this has been established they can be further divided into the "meso" categories of backwater or scour . Finally, pool types are even more detailed into subcategories organized under the meso categories of backwater or scour. Total area of micro habitats identified by Bisson et al. 1981 and total number of fish in the sample reaches (Channel types) will be estimated. Visual estimates of area will be made for all micro habitats within the stream reach, and visual estimates of fish numbers will be made for every fifth habitat feature. The visual estimates will all be calibrated against measured data.

Habitat units will be separated into three levels of hierarchy. First is macro-units; pools, glides, riffles, and side channels. The second is meso-units, which are sub-divisions of macro-units. Microunits further divide channel units (Table 1). Habitat units will be identified by first giving them a channel type or reach number. All habitat units will be visually identified and recorded. A twenty percent sample will be used, and every fifth habitat feature will be measured. These habitat units will be measured with a tape. Area (sq. meter) will be derived from length and width measurements of the feature.

To address the problem of micro habitat features differing substantially due to flow levels, total residual pool volumes will be measured. It is recognized that habitat units can vary significantly in relation to streamflow. So in addition to the three levels of habitat unit identification, residual pool depth measurements will also be taken. the concept was originally described by Bathhurst (1981) and is a excellent method of measuring pool depth independently of discharge. Essentially, the residual pool depth is equal to the difference between the pool's maximum water depth and the tail crest water depth.

Juvenile salmonids will be counted using snorkel surveys of twenty percent of the habitat units. The starting point will be randomly selected, and subsequent habitat features will be surveyed at regular intervals. Most cases a single diver will take counts in each habitat unit. In the case of larger habitat features, the habitat will be segregated into smaller units and sampled. Total fish counts are assumed, though some fish will be missed, so counts will be considered minimum.

The third set of data measured will be substrate data. This is important in PWS where available spawning area is often used as an indication of salmonid production potential. To reduce variability of induced by observer bias, Wolman pebble counts (Wolman 1954) will be used to determine

distribution of substrates and size distribution by channel type, and to assess substrate condition and its relation to fish habitat needs. To be considered as spawning habitat either a portion of, or the entire habitat unit has to meet four criteria. That is average particle size of the substrate not greater than 125 mm. Silt and sand combined can not exceed thirty percent of the substrate composition. The substrate can not be highly compacted or cemented together ( the gravel must be readily moved by twisting ones foot). Last, those areas that meet the previous three criteria must be minimally two square meters in size. The percent area with the habitat unit that meets all four criteria will be estimated. For those substrates that meet the spawning criteria, an estimate of percent composition for five sediment sizes (silt <0.06mm, sand 0.06 to 2mm, gravel 2 to 64mm, small cobble 64 to 128 mm, and large cobble 128 to 256mm).

Frequency distributions of macro, meso, and micro habitats will be calculated for all channel types sampled. Also, percent area of the habitat features by channel type will also be calculated. Multi variate analysis of variance testing (MANOVA) (SAS 1988) will be used to test the effect of channel type on the areas of in-stream habitat attribute features. Juvenile salmonid densities will be compared between habitat units. Single analysis of variance testing will be used to test the effect of habitat units (macro, Meso, and micro) on the salmonid densities.

## cy AssStance

Contract for charter vessel will be required.

#### D. Location

The survey area within which study sites will be located in at least ten watershed randomly selected within oil spill zone in Prince William Sound. Focus will be on the islands within western Prince William Sound and the mainland west of Long Point.

## SCHEDULE

#### sks forÆY 96

Field inventory would occur during summer of 1996. Data input and analysis October 1996 through February 1997. Report preparation and acquisition parcel comparison analysis would be completed by June 1997.

Start-up to June 1:	Arrange logistic (charter boat contract, equipment, hire temporaries)
June 1 - August 31:	Conduct survey of 10 watersheds
September 1 - December 1:	Analysis of stream data
April 1997:	Annual report on FY 96 work

## nd Endpoints

The channel type habitat relations project will provide a GIS based tool allowing comparative evaluations of streams throughout the oil spill affected area. The Alaska Region Channel Type system is a unique system developed for classification of streams on National Forest system lands in Alaska. The streams of the *EVOS* areas have been classified using this system under a previous project (Stream Classification Study, Project Number 99505, Part C). The staff of the Chugach National Forest have extensive knowledge of this system, and are familiar with the techniques to estimate basin-wide fish habitat.

## oject Réports

Data analysis will be complete by February 1997. Draft report should be complete by June 1996. Peer review complete by August of 1996. With final report complete by end of FY 97, October 1997.

## COORDINATION AND INTEGRATION OF RESTORATION EFFORT

This project is an extension of previous projects designed to provide comparative evaluations of fish habitat within comparative evaluation parcels.

## ENVIRONMENTAL COMPLIANCE

No environmental assessments are required.

#### PERSONNEL

Project Co-leader: Steve Zemke received his B.S. in fisheries science from the University of Idaho in 1973. Steve has worked for the Forest Service as a fisheries biologist in Regions 4, 6, and 10. He has extensive experience in planning, inventory, and enhancement of anadromous fish resources. From 1985 to 1991, he was fisheries program leader on the Ketchikan Area of the Tongass National Forest. One of the primary accomplishments of his tenure was the implementation of the Alaska, Region Channel type classification on the Ketchikan Area. He co-authored the 1994 Stream Channel Type Classification report for the EVOS area.

Project Co-leader: Robert Olson received his M.S. in aquatic ecology from the State University of New York in 1983. Robert worked for the USFWS on Kodiak Island performing research on red salmon population dynamics. He has extensive Alaskan field experience, and has a detailed background in statistical analysis. He has also worked as the Chugach National Forest Fish Habitat Relationship Program Coordinator. He is extremely familiar with PWS and the Alaska Region channel type classification system. He co-authored the *EVOS* Stream Channel Classification report for the oil spill area.

Robert Olson and Steven Zemke will each dedicate ten months to the project and will act as co-project

leaders and joint authors of the final project report. Field work will largely be accomplished by two crews of bio-technicians each composed of two fisheries biologists each. The four fisheries biologists will work for a total of four months each. One bio-technician will work an additional 3 months

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Macro-units	Meso-units	Micro-units
Pool (PL) - water is slower and deeper than reach average; gradient less than 1%.	Backwater pool (Bw) -associated with an obstruction; flow diverges from thalweg.	Dammed pool (Dm) -upstream of an obstruction: partially or completely blocks flow.
		Eddy pool (pp) -downstream of a completely blocking obstruction.
	Drawdown pool (Dd) -associated with thalweg:flow rapid at entrance, slow in middle and accelerates at the exit.	Plunge pool (pp) -down-stream of a completely blocking obstruction.
		Lateral scour pool (Lsc) -along channel bank; may be associated with a channel constriction.
	Riffles (Rf) -streambed disturbs surface: less than 10% of streambed breaks surface.	Mid-channel scour pool (Msc) - channel constriction causing scour near or mid channel.
		Riffles (r) -gradient from 2 to 4%: flow consistent over reach.
		Low gradient boulder riffle(Lbr) - gradient from 1 to 3%; areas of low velocity and larger substrate protruding from surface.
		Slip-face cascade (ps) -series of small falls and pools; pool lengths are less than on channel width.
		Step-pool cascade (ps) series of small falls and pools; pool lengths are less than one channel width.
	Rapid/Falls (Fls) -high velocity, high turbulence; gradient greater than 5%.	,
	Glide (Gl) -water depth less than channel width.	
Glide/run (GR) -flow slower and deeper than a riffle, but greater than a pool. Surface smooth.	Run (Rn) -water depth close to or greater than channel width.	
Side channel (Sc) -sub-channel of the main channel; water either flowing or standing; may be disconnected at low flows; source of water is the main channel. May be sub-divided into meso- or micro-units.		

Table 1.	Habitat	units and	descript	ions used	in channel	type survey	(From B	rvant et al.	1992)
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#### BIBLIOGRAPHY

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October 1, 1995 - September 30, 1996

		Authorized	Proposed						
Budget Category:		FFY 1995	FFY 1996						
Personnel			\$112.9						
Travel			\$1.4						
Contractual			\$77.5						
Commodities			\$0.5						
Equipment			\$3.4		LONG F	RANGE FUNDIN	<b>G REQUIREME</b>	NTS	
Subtotal		\$0.0	\$195.7	Estimated	Estimated	Estimated	Estimated	Estimated	Estimated
General Administratio	n		\$22.4	FFY 1997	FFY 1998	FFY 1999	FFY 2000	FFY 2001	FFY 2002
Project Total		\$0.0	\$218.1	\$40.3					
Full-time Equivalents	(FTE)		3.1						
				Dollar amount:	s are shown in	thousands of c	Iollars.		
Other Resources									
				·					

Pers	onnel Costs:		GS/Range/	Months	Monthly	I	Propose
PM	Name	Position Description	Step	Budgeted	Costs	Overtime	FFY 199
	Steven Zemke	Project Leader	GS11/10	7.0	4,838		33.9
	Robert Olson	Fisheries Biologist	GS11/4	4.5	4,478		20.2
	Vacant	Fish Tech	GS7	9.0	2,180		19.6
	Vacant	Fish Tech	GS5	16.0	2,080		33.3
٠	R. Thompson	Program Manager	GS13	1.0	5,928		5.9
l							0.0
							0.0
H							0.0
l							0.0
	1					1	0.0
8							0.0
							0.0
		Subto	tal	37.5	19,504	0	
Tho	se costs associated with	program management should be indicated by pla	scement of an *.		F	Personnel Total	\$112.9
Trav	el Costs:		Ticket	Round	Total	Daily	Propose
PM	Description		Price	Trips	Days	Per Diem	FFY 199
ł	Alaska Railroad transfer	from Portage to Whittier	17	25			0.4
ł	Alaska Railroad vehicle	transfer between Portage and Whittier	200	5			1.0
1							0.0
							0.0
I							0.0
1							0.0
							0.0
							0.0
							0.0
1							0.0
l							0.0
				l			0.0
Tho	se costs associated with	program management should be indicated by pla	icement of an *.			Travel Total	\$1.4

## October 1, 1995 - September 30, 1996

1996	Project Number: Project Title: Channel type habitat relationships Agency: USFS	FORM 3B Personnel & Travel DETAIL
		DETTTE

October 1, 1995 - September 30, 1996

Contractual Costs:				Proposed
Description				FFY 1996
5 fixed wing aircraft	to PWS @ \$500/trip	**************************************		2.5
Boat charter - 50 day	vs @ \$1,550			75.0
1				
				A77 F
vvnen a non-trustee organ	ization is used, the form 4A is required.	C(	ontractual lotal	\$77.5
Commodities Costs:			. ,	FIODOSED
Channel type relation	ship forms printing			0.5
		Com	modities Total	\$0.5
	Project Number:			ORM 3B
1006			Co	ntractual &
1330	Project litle: Channel type habitat relationships		Co	mmodities
	Agency: USFS			DETAIL
		<b>1</b>		

## **1996 EXXON VALDEZ TRÜSTEE COUNCIL PROJECT BUDGET** October 1, 1995 - September 30, 1996

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New	Equipment Purchases:		Number	Unit	Proposed
Desc	ription		of Units	Price	FFY 1996
	Dry suits		2	1,000	2.0
	Diving gear		2	100	0.2
	Hip boots		12	100	1.2
					0.0
					0.0
		• •			0.0
					0.0
					0.0
					0.0
					0.0
					0.0
					0.0
Thos	e nurchases associated with r	enlacement equipment should be indicated by placement of an B	New F	winment Total	0.0
Friet	ing Equipment Usage	eplacement equipment should be indicated by placement of an n.	New L	Number	Inventory
Desc	riotion			of Units	Agency
	1996	Project Number: Project Title: Channel type habitat relationships Agency: U <u>S</u> FS		F	ORM 3B quipment DETAIL

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## Kenai Habitat Restoration & Recreation Enhancement Project

**Project Number: Restoration Category: Proposer:** Lead Trustee Agency: **Cooperating Agencies: Duration:** Cost FY 96: Cost FY 97: Cost FY 98: Geographic Area: **Injured Resource/Service:**  96180 Revised General Restoration ADNR/ADFG ADNR/ADFG DOI Three Years 560.6 879.6 759.6 Kenai Peninsula Pink salmon, sockeye salmon, Dolly Varden, commercial fishing, subsistence, recreation & tourism.

## 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

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.

## 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.

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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. Summary of Major Hypotheses and Objectives

The project's major objective is to restore injured fish habitat and to establish public use patterns (i.e., sportfishing, camping, etc.) within the Kenai River watershed that are compatible with habitat protection for injured resources such as intertidal marshes, pink and sockeye salmon and Dolly Varden. The long

term goal of the project is to protect fish and wildlife habitat, enhance recreation and preserve the functions and values that the riparian habitat contributes to the watershed.

Construction of elevated, grated boardwalks; exclosures; floating docks; fencing; signs; and well designed public use facilities will facilitate restoration and protection of riparian vegetation, stabilize streambanks, and maintain/enhance fish habitat by mitigating the effects of human use.

- . Recreational enhancements will promote continued use of project sites while allowing riparian habitat to recover.
- . Construction of educational and interpretive displays will aid restoration efforts and provide the public with information that will help them to use the river in a manner that protects and sustains its resources.

#### D. Completion Date

The project is conceived to be a multi-year effort that will build on results determined from the monitoring of the previous year's work. Projected timeline: FY 96 to FY 99.

## COMMUNITY INVOLVEMENT

It is intended that the project be fully integrated with on-going agency recreation management, ermitting 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. Whenever feasible, volunteers will be recruited from the local communities to work on the project.

FY 96 BUDGET:

Personnel	228.8
Travel	15.8
Contractual	253.0
Commodities	11.0
Equipment	0.0
Subtotal	508.6
Gen. Admin.	52.0
Total	560.6

## **PROJECT DESIGN**

## A. Objectives

- 1. Review existing information on:
  - a) human impacts to the riparian zone on public lands in the Kenai River watershed in order to select project sites.
  - b) recreational use patterns and infrastructure support in the Kenai River watershed.
  - c) appropriate restoration and/or access and use of enhancement technique(s).
- 2. Select and design the appropriate revegetation, rehabilitation and/or enhancement project for each site.
- 3. Develop an evaluation process to prioritize project sites and define scope of work.
- 4. Construct projects using a combination of competitively bid contracts and volunteer help, where feasible.
- 5. Verify compliance with restoration designs and evaluate construction.
- 6. Implement a monitoring program to assess restoration and use of project sites.
- 7. 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.

During the first year of the project, we will review information from existing studies of the Kenai River on fish habitat, shoreline characteristics, streambank damage, streambank rehabilitation, land ownership, public use trends, development threats, habitat protection/recreation enhancements, infrastructure and public access. On-going and completed restoration projects on the river will also be inspected. This information will be supplemented with personal observations of ADNR and ADF&G staff who have expert knowledge of the river and its use by the public. These data will be used to document the existing condition of potential sites and used later as a baseline for monitoring project success. The data will also be used to develop an evaluation and ranking system to prioritize projects.

Although these data are useful for a broad, area-wide approach, they are not adequate for specific site design. Consequently, once a preliminary list of sites is selected, on-site verification/assessment will be carried out. The final list of project sites will reflect the results of these assessments. 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. The project intends to utilize volunteers to assist with construction and installation. The DNR Parks and Outdoor Recreation Division has an established network of contacts with volunteer organizations on the Kenai Peninsula. DNR has successfully used volunteers in trail construction and park maintenance efforts. Each site plan will include a maintenance element. Maintenance may include watering, fixing fences, replacing signs and/or repair of habitat enhancements.

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 success or failure of each project. 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. Once it is determined that restoration/enhancement is proceeding on an acceptable course and rate, monitoring measurements will be taken less frequently. 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.

## C. Contracts and Other Agency Assistance

All components of the project will be carried out by personnel from ADF&G and ADNR. Construction work will be carried out by contractors on an "as-needed" basis depending on the project design. Volunteers supervised by agency staff will assist in the installation of prefabricated structures and in routine maintenance.

## D. 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.

## SCHEDULE

## A. Measurable Project Tasks for FY 96

Startup to April 15:	Acquire and review existing data on Kenai River, Develop implementation strategy, i.e., applicability of techniques to
	different site conditions,
	Review planned, on-going and completed restoration projects in the Kenai River watershed,
	Develop site evaluation, ranking and priorization system,
	Conduct pre-construction site surveys, assessments and data collection,
	Develop design plans for restoration, enhancement and education components,
	Apply for Title 16, Parks and COE permits for first priority sites,
	Conduct public scoping meetings and resource agency meetings and prepare environmental compliance documents,
	Harvest and store plant materials,
	Organize volunteer support.
April 16 to May 15:	Review comments and revise environmental compliance documents,
	Respond to permitting issues and secure construction permits,

	Conduct construction work, e.g., floating docks, needed below the OHW elevation on first priority sites.
May 16 to July 15:	Construct bank stabilization and revegetation projects, construct boardwalks, fencing, signage, etc
	Design and put up signs and information displays.
July 16 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 16 to September 30:	Continue monitoring,
- ·	Prepare annual report.

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

#### Startup to November 1, 1995

Review existing information on:

- a) human impacts to the riparian zone on public lands in the Kenai River watershed in order to select project sites.
- b) recreational use patterns and infrastructure support in the Kenai River watershed.
- c) appropriate restoration and/or access and use enhancement technique(s).

November 1, 1995 to April 15, 1995

Select and design the appropriate revegetation, rehabilitation and/or enhancement project for each site.

Develop an evaluation process to prioritize project sites and define scope of work.

#### April 16, 1995 to July 15, 1995

Construct projects using a combination of competitively bid contracts and volunteer help, where feasible.

Verify compliance with restoration designs and evaluate construction.

#### July 16, 1995 to August 15, 1995

Implement a monitoring program to assess restoration and use of project sites.

Design and construct educational and interpretive signs and displays.

### August 16, 1995 to September 30, 1995

Continue monitoring,

Prepare annual report.

## FY 96 and Beyond

Continuing work will include primarily permitting, construction and monitoring. Environmental compliance and public coordination efforts will also continue.

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## C. Project Reports

An annual report will be prepared detailing results of the previous year's efforts including monitoring. This report will be submitted to the Chief Scientist in the Fall of each year.

## 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.

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## ENVIRONMENTAL COMPLIANCE

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The revegetation and education elements of the project are categorically exempt from formal documentation in an Environmental Assessment or Environmental Impact Statement according to NEPA guidelines. In-stream construction of floating docks, boardwalks, access stairs or other in-stream structures will require an Environmental Assessment and a Title 16 permit. All permits will be obtained prior to commencement of on-site improvements.

## 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.

Mark Kuwada, Project Leader Division of Habitat and Restoration AK Department of Fish & Game 333 Raspberry Road Anchorage, AK 99518-1599 (907) 267-2277 FAX (907) 349-1723

Lance Trasky, Project Manager Regional Supervisor Division of Habitat and Restoration AK Department of Fish & Game *333 Raspberry Road Anchorage, AK 99518-1599 (907) 267-2335 FAX (907) 349-1723

#### **Project Leader**

TBD - DNR will appoint a project leader with the following qualifications:

B.S. and graduate degree(s) in biology, zoology and/or fisheries.

Extensive experience in field biology, permitting, design and construction of restoration projects and in coordinating departmental policy with other state and federal resource agencies. The project leader will have a working knowledge of the natural resources and human uses of the Kenai River watershed.

Marty K. Rutherford, Project Manager Deputy Commissioner Alaska Department of Natural Resources 3601 C Street, Suite 1210 Anchorage, AK 99503 (907)-762-2483 FAX (907) 562-4871

## REFERENCES

-10

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.

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1996 EXXON VALDEZ TRUE COUNCIL PROJECT BUDGETOctober 1, *1995 - September 30, 1996

	Authorized	Proposed		PROPOSED I	FFY 1996 TRUS	STEE AGENCIE	ES TOTALS	
Budget Category:	FFY 1995	FFY 1996	ADEC	ADF&G	ADNR	USFS	DOI	NOAA
				\$291.4	\$383.1			
Personnel	\$0.0	\$228.8						
Travel	\$0.0	\$15.8						
Contractual	\$0.0	\$253.0						
Commodities	\$0.0	\$11.0						
Equipment	\$0.0	\$0.0	<b>i</b>	LONG F	RANGE FUNDIN	IG REQUIREN	IENTS	
Subtotal	\$0.0	\$508.6	Estimated	Estimated	Estimated	Estimated	Estimated	Estimated
General Administration	\$0.0	\$52.0	FFY 1997	FFY 1998	FFY 1999	FFY 2000	FFY 2001	FFY 2002
Project Total	\$0.0	\$560.6	\$879.6	\$759.6	\$0.0	\$0.0	\$0.0	\$0.0
	3							
Full-time Equivalents (FTE)	0.0	3.1						
			Dollar amoun	t <mark>s</mark> are shown ir	n thousands of d	ollars.		
Other Resources	\$0.0	\$0.0	\$0.0	\$0.0	\$0.0	\$0.0	\$0.0	\$0.0
1. Construction of boardwalks to a impacted shorelines on public land designated for restoration and ent	minimize user ir ds including the hancement.	npact, 2. Active construction an	in revegetation of	f degraded sho signs, fencing	AUG 1	Iands, and 3. F anisms to char	Passive restorational international recreational	ion of I use of sites
1996 Prepared:	Project Num Project Title: Lead Agency	ber: 9619 Kenai Habit y: AK Dept. c	30 at Restoratio of Natural Res	n and Enhan sources	cement		¶ ∎ F	FORM 2A PROJECT DETAIL

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

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Budget Category:	Authorized	Proposed EEV 1996						
Suger Subgery.	1111000							
Personnel		\$93.0						
Travel		\$5.3						
Contractual .		\$116.0						
Commodities		\$5.5						
Equipment		\$0.0		LONG R	ANGE FUNDIN	<b>NG REQUIREM</b>	ENTS	
Subtotal	\$0.0	\$219.8	Estimated	Estimated	Estimated	Estimated	Estimated	Estimated
General Administration		\$22.1	FFY 1997	FFY 1998	FFY 1999	FFY 2000	FFY 2001	FFY 2002
Project Total	\$0.0	\$241.9	\$440.0	\$400.0				
Full-time Equivalents (FTE)		1.2						
			Dollar amour	nts are shown ir	n thousands of	dollars.		
Other Resources						[		
Comments:							١	
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	Project Num	her [.]						FORM 3A
1996	Project Title	Konai Hahit	at Restoratio	n and Enhan	cement			AGENCY
		Dent of Nat	ural Resource	as and critical		ĺ		PROJECT
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#### 1996 EXXON VALDEZ TRU

October 1,*1995 - September 30, 1996

Pers	sonnel Costs:		GS/Range/	Months	Monthly		Proposed
PM.	Name	Position Description	Step	Budgeted	Costs	Overtime	FFY 1996
	TBD	Natural Resource Manager II	20	12.0	<b>7,00</b> 0	0	84.0
	TBD	Park Ranger II	16	2.0	4,500		9.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
		Subtotal		14.0	11,500	0	
Tho	se costs associated with progra	am management should be indicated by placer	ment of an *.		Pe	ersonnel Total	\$93.0
Trav	/el Costs:		Ticket	Round	Total	' Daily	Proposed
PM	Description		Price	Trips	Days	Per Diem	FFY 1996
							0.0
	Travel to Kenai to attend mee	tings, conduct site evaluations, inspections,	114	20	20	150	5.3
	supervise and monitor constru	lction and revegetation.					0.0
							0.0
							0.0
							0.0
1							0.0
							0.0
							0.0
							0.0
							0.0
Tho	se costs associated with progra	ment of an *.			Travel Total	\$5.3	
						,	
							FORM 3B
1996 Project Number: Project Title: Kenai Habitat Restoration and Enhancement						*	Personnel
							& Travel
			DETAIL				
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# **1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET** October 1,*1995 - September 30, 1996

Contractual Costs	мания и на		Proposed
Description		······································	EFY 1996
			11,1000
Planning, orga 6 volunteers/2	nization and coordination of volunteers, including recruitment and on site support. Assumes a min. of mos, housing, and monthly stipend.		15.0
Alaska Consel pool of superv	rvation Corps, on site supervision of volunteers, coordination, site logistics. (This provides a reliable isory personnel with construction experience necessary for efficient use of volunteer labor pool)		24.0
Signage provid	ding information on closures, alternative use, habitat restoration.	l	10.0
Passive reveg	etation; largely labor intensive requiring minimal material and contractual costs.		10.0
Consultant, pla	ant materials center for preliminary consultation, assessment and monitoring as needed.		10.0
Boardwalk Ins	tallation as needed on approximately 300 linear feet with appropriate river access provided to reduce	i	
river bank imp	act. (Estimate is based upon 300 feet @ \$150.00 per linear foot, actuals may vary due		45.0
to site specific	considerations.)		
Equipment rer	ital.		2.0
M/hon a non tructor	organization is used the form AA is required	Contractual Total	\$116.0
	s organization is used, the form 4A is required.		
Commodities Cos			Proposed
Description			FF1 1990
Field equipme to utilize existi	nt as needed, may include: shovels, picks, fencing supplies, and other hardware. Every effort will be mad ng supplies currently on hand with Division of Parks and other agencies working in the Kenai area.	е	5.0
Office Supplie	s (includes paper, toner cartridges, data cartridges, mailing labels, large mailing envelopes etc.)		0.5
	e (meladee paper, terrer earmagee, aala earmagee, maning labele, large maning errerepee etc.)		0.0
	Coi	nmodities Total	\$5.5
	Project Number:	F	ORM 3B
1006	Project Titler, Kensi Habitat Destaration and Enhancement	Co	ntractual &
1330	Agency AK Dent of Network Descuration and Enhancement		ommodities
	Agency: AN Dept. of Natural Resources		DETAIL

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October 1,#1995 - September 30, 1996

New Equipment Purchase	S:	Number	Unit	Proposed
Description		of Units	Price	FFY 1996
				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	d with replacement equipment should be indicated by placement of an R.	New Ec	uipment Total	\$0.0
Existing Equipment Usage	Э:		• Number	Inventory
Description			of Units	Agency
			l r	
	Project Number:			
1996	Project Title: Kenai Habitat Restoration and Enhancement		<b>f c</b>	
<b>1</b>	Agency: AK Dept. of Natural Resources			DETAIL
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# 1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET October 1,[#]1995 - September 30, 1996

Budget Category:	Authorized	Proposed FFY 1996						
		11110000						
Personnel		\$107.5						
Travel		\$5.3			ana sa			
Contractual		\$137.0						
Commodities		\$5.5						
Equipment		\$0.0		LONG H	ANGE FUNDIN	IG REQUIREM	ENTS	
Subtotal	. \$0.0	\$255.3	Estimated	Estimated	Estimated	Estimated	Estimated	Estimated
General Administration		\$25.7	FFY 1997	FFY 1998	FFY 1999	FFY 2000	FFY 2001	FFY 2002
Project I otal	\$0.0	\$281.0	\$400.0	\$320.0				
		4.4						
	I	1.4	Delles emour	te ere ekenin i	a that says and a st	dellere		
Other Resources			Dollar amoul	its are shown in	n thousands of	I I I I I I I I I I I I I I I I I I I		
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1996	Project Title	Kenai Hahil	tat Restoratio	n and Enhan	rement			AGENCY
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	Ngeney. AN		and Game					DETAIL
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## 1996 EXXON VALDEZ TRUS . ___ COUNCIL PROJECT BUDGET October 1, 1995 - September 30, 1996

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Pers	ersonnel Costs:		GS/Range/	Months	Monthly		Proposed
PM	Name	Position Description	Step	Budgeted	Costs	Overtime	FFY 1996
	TBD	Project Manager	24	1.0	7,500		7.5
	TBD	Habitat Biologist III	18	12.0	6,500		<b>7</b> 8.0
	TBD	Habitat Permitter/Biologist	18	2.0	6,500		13.0
	TBD .	Fish & Game Technician	11	2.0	4,500		9.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
<b> </b>	1	Subtota		17.0	25,000	0	
Tho	se costs associa	ated with program management should be indicated by place	ement of an *.		Pe	ersonnel Total	\$107.5
Trav	el Costs:		Ticket	Round	Total	' Daily	Proposed
PM	Description		Price	Trips	Days	Per Diem	FFY 1996
							0.0
	Travel to Kena	i to attend meetings, conduct site evaluations, inspections,	114	20	20	150	5.3
	supervise and	monitor construction and revegetation.					0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
Tho	se costs associ	ated with program management should be indicated by place	ement of an *.			Travel Total	\$5.3
[							
							FORM 3B
Project Number:						*	Personnel
	1996	Project Title: Kenai Habitat Restorati	on and Enhan	cement		T	& Travol
		Agency: AK Dept. of Fish and Game					
							DETAIL

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October 1,#1995 - September 30, 1996

Contractual Costs:		Proposed					
Description		-FT 1990					
Active revegetation as needed of foot depending on existing site	on approximately 1,000 linear feet of river frontage. (Costs may range from \$50 to \$300 per linear conditions: estimate reflects an average cost of \$100 per linear foot)	100.0					
Passive revegetation; largely lab	Passive revegetation; largely labor intensive requiring minimal materials and contractual costs.						
Signage providing information on closures, alternative use, habitat restoration. Equipment rental.							
When a non-trustee organization is u	ised, the form 4A is required. Contractual Total	\$137.0					
Commodities Costs:		Proposed					
Description		FFY 1996					
Field equipment as needed, may to utilize existing supplies current	y include: shovels, picks, fencing supplies, and other hardware. Every effort will be made ntly on hand with Division of Parks and other agencies working in the Kenai area.	5.0					
Office Supplies (includes paper,	, toner cartridges, data cartridges, mailing labels, large mailing envelopes etc.)	0.5					
	Commodities Total	\$5.5					
1996	Project Number: Project Title: Kenai Habitat Restoration and Enhancement Agency: AK Dept. of Fish and Game	ORM 3B ntractual & mmodities DETAIL					

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**1996 EXXON VALDEZ TRU** E COUNCIL PROJECT BUDGET October 1,[‡]1995 - September 30, 1996

New Equipment P	Jrchases:	Number	Unit	Proposed
Description		of Units	Price	FFY 1996
				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 a	ssociated with replacement equipment should be indicated by placement of an R.	New Ec	uipment Total	\$0.0
Existing Equipme	nt Usage:	· · · · · · · · · · · · · · · · · · ·	Number	Inventory
Description			of Units	Agency
<u>l</u>				
[]				
	Project Number		1 1 1 F	FORM 3B
1996	Project Title: Kenai Habitat Restoration and Enhancement		l E	Equipment
	Agency: AK Dent of Fish and Game			DETAIL
	Agency. An Dept. of Fibit and Game			
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## 1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET October 1, 1995 - September 30, 1996

Budget Category:	Authorized FFY 1995	Proposed FFY 1996						
Personnel Travel		\$28.3						
Contractual	-	\$0.0						
Commodities		\$0.0						
Fauipment		\$0.0			ANGE FUNDIN		ENTS	
Subtotal	\$0.0	\$33.5	Estimated	Estimated	Estimated	Estimated	Estimated	Estimated
General Administration	4010	\$4.2	FFY 1997	FFY 1998	FFY 1999	FFY 2000	FFY 2001	FFY 2002
Project Total	\$0.0	\$37.7	\$39.6	\$41.6				
Full-time Equivalents (FTE)		0.5						
			Dollar amour	nts are shown ir	n thousands of	dollars.		
Other Resources					· · · · ·		[	
1996	Project Numl Project Title: Agency: US	ber: 96180 Kenai Habi Fish & Wildl	tat Restoratio ife Service	n and Enhar	ncement		*	FORM 3A AGENCY PROJECT DETAIL

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October 1, 1995 - September 30, 1996

Per	sonnel Costs:		GS/Range/	Months	Monthiv		Proposed
PM	Name	Position Description	Step	Budgeted	Costs	Overtime	FFY 1996
	TBD	Biologist	GS-11/12	3.0	5,417		16.3
	Emily Dekker-Fiala	Outdoor Recreation Planner	GS-9	3.0	3,998		12.0
							0.0
							0.0
							0.0
							0.0
							0.0
Ì							0.0
							0.0
							0.0
							0.0
	1	I		6.0	0.415		0.0
Tho	se costs associated wit	th program management should be indicated by place	ment of an *	0.01	9,415 Pc	ersonnel Total	\$28.3
Tra	al Coste:	in program management energia be indicated by place	Ticket	Round	Total	) Daily	Proposed
PM	Description		Price	Trins	Davs	Per Diem	FEY 1996
			11100	11103	Duys		0.0
	Travel to Kenai to atte	and meetings, conduct site evaluations, inspections,	110	7	25	178	5.2
	supervise and monitor	r construction and revegetation.					0,0
		<b>.</b>					0.0
							0.0
							0.0
							0.0
						i	0.0
							0.0
							0.0
							0.0
	L						0.0
lho	se costs associated wit	in program management should be indicated by place	ment of an *.			Travel Total	\$5.2
						l	
							FORM 3B
ł	1006	Project Number: 96180				*	Personnel
	1330	Project Litle: Kenal Habitat Hestoratio	on and Ennan	cement		Ť	& Travel
			DETAIL				

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

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Contractual Costs:	Proposed
Description	FFY 1996
When a non-trustee organization is used, the form 4A is required. Contractual Total	\$0.0
Commodities Costs:	Proposed
Description	FFY 1996
Commodities Total	\$0.0
1996 Project Number: 96180 Project Title: Kenai Habitat Restoration and Enhancement Agency: DOI, US Fish & Wildlife Service	ORM 3B itractual & mmodities DETAIL

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# 1996 EXXON VALDEZ TRUSILE COUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

New Equipment Purchases	S:	Number	Unit	Proposed
Description		of Units	Price	FFY 1996
				0.0
				0.0
				0.0
· ·				0.0
	•			0.0
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
				0.0
Those purchases associated	with replacement equipment should be indicated by placement of an R.	New Eq	uipment Total	\$0.0
Existing Equipment Usage	<u> </u>		Number	Inventory
Description			of Units	Agency
,	4			
· · · · · · · · · · · · · · · · · · ·		]		
	Droje st Number, 00100		¥ F	FORM 3B
Project Number: 96180			ΙE	auipment
Project Title: Kenai Habitat Restoration and Enhancement			-	DETAIL
	Agency: DOI, US Fish & Wildlife Service			
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# Coded Wire Tag Recoveries From Pink Salmon in Prince William Sound

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Project Number:	96186 (formerly 95320B)
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 Corp. Valdez Fisheries Development Assoc.
Duration:	4 years
Cost FY 96: Cost FY 97: Cost FY 98: Cost FY99:	\$260,500 \$260,500 \$260,500 \$260,500 \$ 85,000 \$ 85,000 \$ 85,000 \$ 000 \$ 260,500 \$ 000 \$ 0000 \$ 000 \$ 000 \$ 000 \$ 000 \$ 00
Geographic Area:	Prince William Sound WILL be provided later.
Injured Resource:	Pink Salmon

## ABSTRACT

## INTRODUCTION

Pink salmon play a major role in the Prince William Sound (PWS) 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 PWS 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 PWS communities. Exvessel values for this fishery ranged from 10 to almost 40 million through the 1980's.

PWS pink salmon returns originating from brood years subsequent to the March 24, 1989, *Exxon Valdez Oil Spill* (EVOS) 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 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 smallost in the last 30 years and the hatchery return was less than one fifth of expected. Both wild and hatchery returns of 1994 were a significant improvement over the preceding two years.

There is a growing body of evidence which indicates that the EVOS was partially responsible for the weak pink salmon returns to PWS. 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 EVOS adversely affected spawning success and early marine survival in PWS. Mortalities of pink salmon embryos incubating in the intertidal portions of oiled streams in western PWS have been significantly higher than those which incubated in nearby unoiled streams since 1989 (Sharr et. al. 1994a, Bue et al. (in press)). 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. (in press)). These findings may be indicative of heritable genetic damage which has resulted in reproductive impairment among first and second generation fish originating from populations whose fry 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 PWS in 1989 also exhibited reduced growth and survival (Willette and Carpenter, 1994). Because almost all wild and hatchery fry exit PWS through the straits and passages that were most heavily oiled, it is likely that at least portions of almost all pink salmon populations in PWS were damaged as rearing fry and juveniles in 1989. There are presently no data to substantiate any 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 (see Attachment 1) in PWS 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 coincided with the EVOS era. Returns of wild salmon are dominated by the larger returns from the more productive hatchery populations and are therefrore 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 PWS.

This study will provide accurate, 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. Such catch contribution estimates, together with real-time escapement estimates from an Alaska Department of Fish and Game (ADF&G) aerial survey program will be used inseason by fisheries managers to reduce exploitation on wild stocks and target effort on hatchery returns. Post season analyses of tag recovery data will be coupled with escapement data for wild populations to make estimates of total wild returns, which will in turn allow assessment of the effectiveness of various management strategies. Post season analyses will also identify 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. Similar analyses of coded wire tag data funded by the Natural Resource Damage Assessment (NRDA) and Restoration processes have been used to justify time and area fishery closures and effectively reduce exploitation on oiled populations in portions of southwestern PWS in 1990, 1991, 1992, 1993, and 1994.

The results of the coded wire tag recovery project are also critical to the success of an integrated package of Sound Ecosystem Assessment (SEA) studies. The SEA proposal has roots in a broader SEA plan developed by the Prince William Sound Fisheries Ecosystem Research Planning Group (PWSFERPG), a bioregional coalition of PWS scientists, resource managers, resource users, equaculture associations, and communities, formed to "develop an ecosystem level understanding of the natural and man-caused factors influencing the production of pink salmon...in PWS". Many of the SEA projects, such as those falling under the Salmon Growth Component and the Salmon Predation Component are 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 PWS which have been injured and depleted through oil impacts may be over-exploited in the commercial, sport and subsistence fisheries. Population levels of stocks may be reduced below those needed for rapid recovery and in some instances may result in virtual elimination of impacted stocks. In the absence of the information provided to SEA plan, some of the projects under that plan will fail.

## NEED FOR THE PROJECT

- A. Statement of Problem
- B. Rational
- C. Summary of Major Hypotheses and Objectives

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## D. Completion Date

# COMMUNITY INVOLVEMENT

## FY 96 BUDGET

Personnel Travel Contractual Commodities Equipment Subtotal Gen. Admin. Total

## PROJECT DESIGN

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### A. Objectives

Funds which match those contributed by ADF&G, PWSAC, and VFDA will contribute to the completion of the following objectives for the 1995 salmon season in PWS:

- 1. Using undecoded-tag data, provide timely inseason estimates of the temporal and spatial contributions of tagged hatchery stocks of pink salmon to PWS 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 costrecovery harvests in PWS.
- 4. Estimate marine survival rates for each uniquely coded hatchery release group of pink salmon.

#### B. Methods

Personnel policy, purchasing practices, field camp operations, safety procedures, and project administration will be in compliance the ADF&G Division of Commercial Fisheries Manual of Standard Operating Procedures (SOP). Data collection and estimation procedures are similar to those used in NRDA F/S Study #3. These procedures have been thoroughly reviewed by the NRDA peer review process and approved by the Management Team.

#### Tag Recovery

**Commercial and Cost-Recovery Harvests** 

Recoveries will be 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 will be made as fish are pumped from tenders onto conveyor belts at land-based processors located in Cordova, Valdez, Seward, Anchorage, Whittier and aboard a floating processor after each opening. Fish will be sampled by technicians standing beside the belt. Each

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sampled fish will be subjected to a visual and tactile examination for a missing adipose fin. It will never be possible for an observer to census all fish from a tender during the unloading process. However, on occasion, holding tanks in processing plants contain fish from only one tender. In those instances it will be 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 will be 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 will include 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 will be obtained later from fish tickets.

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Heads of adipose-fin clipped fish will be excised, identified with a uniquely numbered cinch tag, and bagged. These heads will then be individually passed through a tag detector machine which produces an audible signal in the event that the head contains a coded wire tag. This procedure yields numbers of undecoded tags in the sample. Heads will then be frozen for subsequent shipment to the ADF&G Coded Wire Tag Laboratory in Juneau (Tag Lab).

#### **Brood Stock Harvests**

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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 will be scanned for tags in order to estimate adjustment factors which can be used to account for the loss of tags from the population. Three assumptions inherent in the use of the brood stock for this purpose are a) it consists solely of fish reared at the hatchery, b) the propensity for a fish to lose a tag is similar for all fish marked at the same hatchery, and c) for a specific tag code, the marking rate in the commercial fishery is the same as that in the brood stock. It is believed that the first of these assumptions is violated at all facilities except at the W. Noerenberg hatchery (Sharr et. al. 1994f). Consequently, a historical average adjustment factor calculated from the brood stock from the W. Noerenberg hatchery is considered an appropriate quantity with which to adjust for tag loss and differential mortality. With respect to the second assumption, tagging practices vary little within a facility, and it is believed that the rate of tag loss and tag-induced mortality are similar for all fish tagged within a hetchery. 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), and some more direct preliminary evidence is discussed by Sharr et al. (1994f).

The adjustment factor for a given year may be defined as that quantity which, when multiplied by the marking rate in returning fish, yields the marking rate at release. The factor is 1.0 when there is no tag loss or differential mortality. The adjustment factor for

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hatchery h,  $a_{\mu}$ , will be estimated as the ratio of sampled fish in the brood stock to the spanded number of fish based on tags found in the sample :

$$\hat{a}_{h} = \frac{s_{h}}{\frac{r}{\sum_{i} x_{i}}},$$
(1)

where

- T = number of tag codes released from hatchery h,
- $\rho_i$  = 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 in 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 (1989-1994 for inseason, 1989-1995 for postseason) average W. Noerenberg adjustment factor will then be used to adjust contribution estimates (Equation 2) if it can be shown that it was significantly greater than 1.0 at the 90% level.

While only the (historical) adjustment factor associated with the W. Noerenberg facility will be used in any contribution estimation, brood stock samples will be taken during hatchery egg-take operations at each of the four PWS pink salmon hatcheries. Technicians, will examine approximately 95% of the fish through visual and tactile means for missing adipose fins. The number of fish sampled will be recorded and when adipose-clipped fish are found, the heads will be 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 will be sent daily to the ADF&G Tag Lab. Data received at the Tag Lab will be logged and tag recovery sampling forms edited a for accuracy and completeness. Samples which affect critical fisheries decisions will be processed first. Tag lab staff will locate and remove tags from heads, decode extracted tags, and enter tag code and sample data into a statewide database accessible to biologists in Cordova. Completed tag recovery data for prioritized samples will be 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 will integrate tag recovery and catch data from the ADF&G fish ticket reporting system to estimate hatchery and wild catch contributions. Contribution estimates are used by fisheries managers to implement the inseason management actions required. Following the fishing season, processing of all lower priority tag recovery samples will be completed by the Tag Lab. All tags recovered throughout the season will be examined a second time to insure that they have been properly decoded. All codes will be validated with a master Pacific States Marine Fisheries Commission (PSMFC) list of codes potentially present in Pacific coast fisheries. Fully edited tag code and sampling data from all samples collected during the season will be forwarded to the Cordova office for final summarization and analyses. A complete historic database of coded-wire tag information from PWS tagging and tag recovery programs will be maintained by the ADF&G Tag Lab, the PSMFC and, the Cordova ADF&G. The ADF&G historic fish ticket catch database is maintained at the ADF&G Juneau headquarters office and in the Cordova area office. All coded wire tagging and recovery data and all fisheries harvest data are freely available from any of these sources.

#### Estimation of Contributions and Survival Rates

Post-season 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,  $C_t$ , will be estimated as:

$$\hat{C}_{t} = \sum_{i=1}^{L} x_{it} \left( \frac{N_{i} \hat{a}_{h}}{S_{i} p_{t}} \right) , \qquad (2)$$

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,
- $p_t = proportion of group t tagged,$

 $a_h = adjustment factor associated with hatchery h, and$ 

L = number of recovery strata associated with common property, costrecovery, brood stock, special harvests and escapement in which tag code t was found.

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

$$\hat{Cu}_{t} = \sum_{i=1}^{U} \left[ N_{i} * \left( \frac{\sum_{j=1}^{S} \hat{C}_{tj}}{\sum_{j=1}^{S} N_{j}} \right) \right], \qquad (3)$$

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,

 $C_{ij}$  = contribution of release coded with tag t to the sampled stratum  $j_i$  and

 $N_j =$  number of fish in *j*th sampled stratum.

When a district-week opening is not sampled at all (an infrequent occurrence), the catch from that opening will be treated as unsampled catch of the subsequent opening in the same district.

An estimate of the contribution of tag group *t* to the total PWS return for 1995 will be obtained through summation of contribution estimates for sampled and unsampled strata. An estimate of the total hatchery contribution to the PWS return will be calculated through summation of contributions over all release groups. A variance approximation for  $\hat{C}_{t}$ , derived by Clark and Bernard (1987) and simplified by Geiger (1988) will be:

$$\hat{V}(\tilde{C}_{t}) = \sum_{i=1}^{L} x_{it} \left[ \frac{N_{i} \hat{a}}{s_{i} p_{t}} \right] \left[ \frac{N_{i} \hat{a}}{s_{i} p_{t}} - 1 \right].$$
(4)

Assuming that covariances between contributions of different release groups to a stratum can be ignored, summation of variance components over all tag codes will provide an estimate of the variance of the total hatchery contribution. Inspection of the formula given by Clark and Bernard (1987) for the aforementioned covariances 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 strate are believed to be small (Sharr et al., 1994d).

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

$$\hat{S}_{t} = \frac{\hat{C}_{t} + \hat{Cu}_{t}}{R_{t}}, \qquad (5)$$

#### where

 $C_r =$  contribution of release coded with tag t to sampled strata,

 $Cu_r$  = 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  $S_{\rm c}$  is given by:

$$\hat{\mathbf{V}}(\hat{S}_{t}) = \frac{\sum_{i=1}^{L} X_{it} \left[ \frac{N_{i} \tilde{a}}{S_{i} P_{t}} \right] \left[ \frac{N_{i} \tilde{a}}{S_{i} P_{t}} \right]}{R_{t}^{2}} .$$
(6)

**Inseason Hatchery Contributions** 

Inseason fisheries decisions which must be made on very short notice require rapid, real time analysis of coded wire tag data. Three inseason estimates of hatchery contributions of pink salmon will be generated for each opening. The first and most timely estimate will be 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 will be discerned by passing their excised heads over a scanner identical to those used by the Tag Lab. The estimate of the catch aboard tenders will be obtained from tender captains or processor operators. In the event that catch estimates cannot be obtained, a simple unweighted average (over sampled tenders) proportion of hatchery fish in the catch will be reported. Estimation using undecoded tags requires that assumptions be made about expansion (1/p) and adjustment (a) factors (see Equation 2). For fishery openings in the western and northern portions of PWS, late run returns from PWSAC facilities are assumed to be the only hatchery contributors. For openings in the Southwostern district, an expansion factor which is a weighted average of all expansion factors associated with tags released at the A.F. Koernig, W. Noerenberg and Cannery Creek hatcheries in 1993, will be used. The weighting scheme depends upon historical contributions of hatcheries to the district in question. A similar weighting scheme for expansion factors will be used for the Coghill and Northern districts and will involve historical contributions associated with the Cannery Creek and W. Noerenberg hatcheries. For openings in the eastern part of the Sound, returns to the VFDA Solomon Gulch facility are assumed to be the only hatchery contributors. With respect to an appropriate expansion factor for these openings, the average of all factors associated

with tags released from the Solomon Gulch facility in 1993 will be used. An average historical (1989-1994) adjustment factor associated with the W. Noerenberg facility will be used for all inseason contribution estimates. These estimates can be made available at any stage of the unloading process, and only require that some sampling has been conducted. The precision of the estimate is, of course, increased as more of the catch is sampled. Such readily available, but less precise estimates will play a significant role in those fishery management decisions that have to be made before the more precise estimates which require exact catch figures and larger sample sizes are available. Calculations of in-season contributions will follow those used to generate post-season results (Equation 2). The second estimator will be identical to the first, except that it will be calculated only after sampling of an opening is completed and after exact tender loads have been reported. The result will be a less timely but more reliable estimate. The third estimator will be less timely still because it will rely on exact catch data and extracted and decoded tags. Use of code-specific expansion factors will, however, provide hatchery-specific contribution estimates and will 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 to be 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 costeffective 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 given us the opportunity to distinguish hatchery and wild fish in commercial harvests with reasonable accuracy and precision, it is not without problems. The pink salmon tagging program in PWS 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.

An alternative mark which circumvents the above drawbacks would be desirable. The most likely alternative to coded wire tags are thermal or chemical otolith marks. 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 PWS hatchery pink salmon releases has been proposed for 1995 (Study 95320C). Recoveries of otolith marks from these releases can begin in 1997.

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 PWS pink salmon which allow researchers to distinguish hatchery stocks from all wild stocks. Genetic marks are a possibility but hatchery parent stocks in PWS originated from wild stocks in the area and are shared by more than one facility, and hence are probably not distinguishable.

#### C. Contracts:

Matching funds from PWSAC and VFDA will be conveyed to ADF&G through cooperative agreements.

#### D. Location:

By aiding restoration through improved fisheries management, this project will benefit wild pink salmon populations in PWS and other segments of the marine and terrestrial portions of the PWS ecosystem which are dependent upon pink salmon. Restoration through improved management will also benefit the salmon fishing fleets comprising those Cordova, Valdez, Tatitlek, New Chenega, Whittier, and Seward, fish processing plants in Cordova, Valdez, Whittier, Seward, Anchorage, Kenai, and Kodiak. The project will employ local residents for data collection activities in fish processing plants located in Cordova, Valdez, Whittier, Seward, Anchorage, Kenai, and Kodiak, and at hatcheries in PWS. The project will also employ residents of Juneau for tag extraction and decoding activities performed by the ADF&G Statewide Tag Laboratory. Permanent ADF&G Biologists stationed in Cordova and biometrics staff stationed in Anchorage will complete data analyses and reports. Goods and services required by the project will be obtained from vendors in the local communities where data are collected.

#### SCHEDULE

- A. Measurable Project Tasks for FY 96
- B. Project Milestones and Endpoints
- C. Project Reports

## COORDINATION AND INTEGRATION OF RESTORATION EFFORT

## ENVIRONMENTAL COMPLIANCE

All sampling activities for this project occur within fish processing plants or fish hatcheries. The ADF&G will coordinate with PWSAC and VFDA and the processing plants at which fish are unloaded with respect to locating samplers within their premises.

## PERSONNEL

The Project Leader (PL) for the project is a permanent full time Fisheries Biologist III (FB III), PWS Salmon Research Project Leader with the Alaska Department of Fish and Game. The PL 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 permanent seasonal Fisheries Biologist II (FB II) will act as the Assistant Project Leader (APL), supervise day to day project operations, maintain data quality, assist in data analyses, and coauthor final reports. The APL 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. Each day, two persons on each crew will scan pink salmon at each processing plant. 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 ADF&G 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 PL, APL or, a project FB I will maintain daily phone contact with project technicians stationed in ports other than Cordova or Valdez and at several remote hatchery locations.

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Copies of data forms from these sites will be faxed to Cordova daily and heads from sampled fish 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's 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 post season to assist with final data analyses and report writing.

## 6. Technical Support:

Tag recovery data forms and heads from tagged fish will be shipped to the Cordova office for logging, sorting, editing, and final shipment to the centralized ADF&G Coded Wire Tag Laboratory in Juneau, Ak. Tag Laboratory personnel will use specialized equipment to detect, extract and decode tags. The Tag Laboratory uses a Honeywell minicomputer with an ULTIMATE operating system and PIC database software to construct, manipulate, and store the PWS data in a statewide coded wire tag database. A copy of the statewide database is also incorporated into a Pacific Coest database maintained by the (PSMFC) in Gladstone, Oregon. Summarized data from the Juneau tag laboratory and summaries of ADF&G fisheries sales receipts (fish tickets) are stored and analyzed on micro-computers in the ADF&G Cordova offices and on a mainframe in the ADF&G headquarters office in Juneau. All inseason and post season data analyses and reporting are completed on micro-computers using RBASE database management, LOTUS spreadsheet, and WORDPERFECT word processing software.

### 1. Resources and/or Associated Services:

This restoration project is designed to facilitate the recovery of wild pink salmon populations in PWS. The project is intended to provide fisheries managers with data pertaining to the composition of the catch. These data will be used for improving the management capabilities in PWS fisheries, thereby reducing the exploitation rate on salmon stocks in western PWS which have already been stressed and depleted through oil impacts. Improved management will enhance the chances that damaged wild pink salmon populations are not reduced below those needed for rapid recovery. The monitoring portion of this project will track the recovery of damaged populations.

#### 2. Relation to Other Damage Assessment/Restoration Work:

The foundations for this project were firmly established in joint feasibility studies which were conducted by ADF&G and non-profit aquaculture associations in PWS beginning in 1986 and extending through 1988. Results of these studies have been summarized by Peltz and Miller (1990), Peltz and Gelger (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 (NRDA) Fish/Shellfish (F/S) 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 (NRDA F/S Study #3). Tag recovery efforts for wild and hatchery pink salmon were funded by damage assessment funds in 1989, 1990, and 1991 (F/S 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). Following the loss of funds for further tagging of hatchery stocks of pink salmon in 1990, the private non-profit aquaculture associations in PWS have continued to tag pink salmon releases at their own expense. Tags applied to pink fry from the four pink salmon hatcheries in PWS in 1993 must be recovered. Prince William Sound Aquaculture Corporation (PWSAC), Valdez Fisheries Development Association (VFDA), and the ADF&G have pooled their resources to come up with approximately half of the funds required to field a full fledged pink salmon tag recovery effort in 1995. The additional funds to complete tag recovery efforts and data analyses are to be provided by the EVOS Trustee Council.

The pink salmon coded wire tag recovery project has complimented several other projects since 1989. Improved escapement estimates for PWS pink salmon from NRDA F/S 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 NRDA F/S Study #4 could not have been obtained without F/S Study #3 which provided coded wire tagged fish of known origin and release timing.

#### C. SCHEDULES

Date(s)	Activity
October 1,1994 February 15,1995	Draft FY 94 report, Draft FY95 DPD
January 15-June 20, 1995	Hire personnel, order supplies, create and test appropriate inseason spreadsheets
June 20-Sept 30, 1995	Tag recoveries in commercial fisheries, cost recovery harvests, and brood stocks. Inseason catch composition estimates by time and area.
January 15, 1996	Draft Report
May 30, 1996	Final Report

#### D. EXISTING AGENCY PROGRAM

The Alaska Department of Fish and Game permanent staff of biologists and biometricians write operational plans and provide overall supervision for this project. The Alaska Department of Fish and Game, PWSAC, and VFDA also provide matching funding for project operations. These funding contributions for the period October 1, 1994 through September 30, 1995 are as follows:

#### ADF&G

PWSAC	\$ 100.0K
VFDA	\$ 26.2K

In addition, data and personnel from ongoing ADF&G fisheries catch and escapement monitoring and management programs will be used in conjunction with results of this study to make fisheries catch contribution estimates and formulate stock specific management strategies.

#### E. ENVIRONMENTAL COMPLIANCE/PERMIT/COORDINATION STATUS

#### F. PERFORMANCE MONITORING

The ADF&G, Commercial Fisheries Management and Development Division (CFMD), Region II, Regional Research Supervisor supervises the PL, the permanent full time Fisheries Biologist III PWS Salmon Research project Leader for CFMD. The APL is an eleven month permanent season ADF&G employee and is supervised by the PL. The APL has supervisory authority over the Fisheries Biologist I's who in turn has supervisory authority over Fish and Wildlife Technicians. A Biometrician assigned to assist the PL and APL is supervised by an ADF&G, Commercial Fisheries Management and Development Division, Region II, Biometrician II. The PL and the project Biometrician coordinate through the regional Biometrician II, the Regional Biometrician III, and the Regional Research Supervisor. The various levels of supervision are depicted in Figure 1. The PL and APL have equal knowledge of all aspects of this project and can exchange roles in the event of a personnel change. In addition, the Project Fisheries Biologist | has sufficient knowledge and experience with the project that she could be promoted to the APL position and trained in data analysis and report writing tasks very quickly. Biometrics responsibilities are interchangeable between the Biometrician 1 and the Biometrician II. Technician III crew leaders with the project can be replaced in the short term by the Fisheries Biologist I. Several Technician II's have been with the project for more than one season and qualify as easily trained replacements.

Sampling materials, data forms, and sampling equipment will be purchased or shipped to Cordova from the ADF&G Statewide Coded Wire Tag Laboratory no later than June 1, 1995. Sampling protocol, data forms, data recording procedures and conventions, data editing procedures, and data transmission procedures are all in accordance with statewide standards established by the ADF&G Statewide Coded Wire Tag Laboratory. Data standards adopted by the ADF&G Statewide Coded Wire Tag laboratory are in accordance with those used by the PSMFC. All data are edited immediately upon completion of sampling and are edited twice more by Statewide tag laboratory personnel.

Data sheets will be edited and logged and heads from these samples will be scanned immediately for estimates of the number of undecoded tags. Preliminary estimates of wild stock catch contributions will be made from this undecoded-tag data and these estimates will be made available to fisheries managers as soon as possible. Samples from district-period strata which are at the centre of potentially controversial management decisions will be given priority as far as reporting and analysis are concerned. Assessment of bias in the sampling of tender loads will be conducted when the opportunity for an independent total census arises. Data sheets and heads and copies of the data log will be shipped to Juneau for tag extraction on the day they are collected. Tag laboratory personnel cross check all samples received with the accompanying copy of the data log and work overtime if necessary to insure that data editing, entry, tag extraction, tag decoding, and data transmission back to Cordova are completed within 36 hours of the time of sample receipt. Project biologists and biometricians in Cordova complete data analyses of decoded tag data and use this data to verify and update preliminary catch contribution estimates based on undecoded tag data. Project biologists will visit each sampling port a minimum of once every two weeks to answer questions, and provide quality control supervision.

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Figure 1. Organizational chart of project personnel and related ADF2g CFND Region II supervisory and biom trics staff.

Following the fishing season processing of all lower priority tag recovery samples will be completed by the coded wire tag lab. In addition, all data collected through the season are edited again for quality control, and all tags recovered throughout the season will be examined a second time to insure that they have been properly decoded. All codes will be validated with a master Pacific States Marine Fisheries Commission (PSMFC) list of codes potentially present in Pacific cast fisheries. Fully edited tag code and sampling data from all samples collected during the season will be forwarded to the Cordova office for final summarization and analyses. A complete historic database of coded-wire tag information from PWS tagging and tag recovery programs will be maintained by the ADF&G statewide coded wire tag laboratory, the PSMFC and, the Cordova ADF&G. The ADF&G historic fish ticket catch database is maintained by the ADF&G at the Juneau headquarters office and in the Cordova area office. All coded wire tagging and recovery data and all fisheries harvest data are freely available from any of these sources.

## G. COORDINATION OF INTEGRATED RESEARCH EFFORT

The monitoring, research and restoration objectives of this project are integral to the success of broader ecosystem research and restoration effort described in part by the Sound Ecosystem Assessment (SEA) plan. The SEA plan is a multi-disciplinary program designed to develop an understanding of the regulatory mechanisms which control the state of the PWS ecosystem. In its first year it has and will evaluate the interactions of pink salmon and herring with other components of the ecosystem. coded wire tagmarked fish will provide a valuable tool for examining interactions between wild and hatchery salmon during their early marine residencies. The Salmon Growth component of SEA will utilize coded wire tag-marked juvenile pink salmon to (1) evaluate habitat overlap between hatchery and wild salmon, (2) compare size composition of wild and hatchery salmon in mixed stocks, and (3) develop a tagging program to estimate juvenile salmon mortality within PWS and in the Gulf of Alaska. The Salmon Predation component of SEA will utilize coded wire tag-marked juvenile salmon to determine if predators have a preference for wild versus hatchery fish. The program is also linked to other studies such as the Pink Salmon Egg and Alevin Mortality Project and the Otolith Mass Marking Project.

This project will integrate tender fleet tracking, processor plant logistics, and crew scheduling with existing ADF&G salmon port sampling projects. Local aquaculture associations (PWSAC, VFDA) provide all tagging, fry release, sales harvest, and broodstock data necessary for data analysis. Aquaculture associations also provide room, board, and logistics support for broodstock samplers at their hatcheries. Air charter and boat transportation required to get samplers to remote locations in PWS will be shared with other projects having similar needs.

## H. PUBLIC PROCESS

The general public has been involved in the development and evolution of the coded wire tag program in PWS since its inception in 1986 as a cooperative effort between ADF&G and the PWS area private non-profit (PNP) aquaculture associations. These PNP's, operated by a broad constituency of commercial, sport, personal use, and subsistence fishers and community representatives, review coded wire tag project plans and results annually before approving subsequent funding. Operational plans and results of the coded wire tag program are also reviewed periodically by the PWS Regional planning team as well as interested fishing industry groups. As part of the Trustee Council NRDA and Restoration process the code-wire tag recovery project has also been subject to extensive peer review and annual public review and comment. Results of the coded-wire tag project have been presented at the March 1993 Oil Spill Symposium sponsored by the Trustee Council, the 1993 and 1994 Pink and Chum Workshop, and at the annual Spring meetings of the PWSAC board of directors in 1993 and 1994.

## I. PERSONNEL QUALIFICATIONS

Fisheries Biologist III Project Leader - To be announced.

Fisheries Biologist II Assistant Project Leader - Renate Riffe

Renate 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 Renate Riff has worked on the coded wire tag project as an FBII 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 ADF&G, 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 ADF&G, 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 LynnCanel sockeye salmon, by scale pattern analysis, developed a computer model which simulated migratory timing of salmon escapements, and evaluated truncated escapement counts.

Fisheries Biologist I - Seawan Gehlbach

Seawan Gehlbach has a Bachelor of Science in biology from the University of New Hampshire (1992). Ms. Gehlbach has worked on the coded wire tag project as an FBI for the past two fishing seasons. Her responsibilities include hiring and supervising 20

Fish and Wildlife Technician II's that sample in eight ports around PWS. In the absence of a project FBI this previous season, she was also responsible for the duties of the current APL, and produced inseason data analysis for management staff and post season data analysis for the annual coded wire tag reports. Prior to her current position with ADF&G, she worked for Sport Fish Division in Juneau, as a short torm Fish and Wildlife Technician II; her duties included collecting coded wire tag data and catch information for the sport fishery. Ms. Gehlbach has also worked for the Douglas Island Pink and Chum (DIPAC) hatchery in Juneau as a field observer, and later in the hatchery as a member of the incubation and broodstock collection crews.

#### **Biometrician I - David Evans**

David 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 (Ontarlo, 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 PWS. 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 NRDA F/S Study #3, the 1992 Restoration Study 60C, and the 1993 Restoration study 93067.

J. BUDGET (attached)

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Figure 1: Fishing districts and hatcheries of Prince William Sound, Alaska

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

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		Authorized	Proposed						
Budget Category		FFY 1995	FEY 1996						
Budget Outegoly.		111 1333	1111000						
Personnel			\$114.5						
Travel		i	\$12.7						
Contractual			\$100.6						
Commodities			\$2.9						
Equipment			\$0.0		LONG F	RANGE FUNDIN	G REQUIREME	NTS	
Subtotal		\$0.0	\$230.7	Estimated	Estimated	Estimated	Estimated	Estimated	Estimated
General Administration	on		\$24.2	FFY 1997	FFY 1998	FFY 1999	FFY 2000	FFY 2001	FFY 2002
Project Total		\$0.0	\$254.9	\$256.9	\$256.9	\$85.0	\$0.0	\$0.0	\$0.0
Full-time Equivalents	(FTE)		2.2						
				Dollar amount	s are shown in	thousands of d	ollars.		
Other Resources								`	
Comments: This is	s a cooperativ	e project betwe	en the Trustee	council, Alaska	Department of	Fish and Game	, Prince William	n Sound Aquac	ulture
Corporation (PWSA)	C) and Valdez	<b>Fisheries</b> Develo	opment Associa	ation (VFDA).	following is a b	reakout of fund	s provided by e	each entity:	
Trustee Council		\$254.9 k							بية
ADF&G		\$80.0	k						ſ
PWSAC		\$100.0	k	•					
VFDA		\$26.2	k						,
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		Project Num	ber: 96186						FURM 3A
1996		Project Title	· Codod \//:	Tag Pasar	rias from Bi	nk Salman P			AGENCY
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**1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET** October 1, 1995 - September 30, 1996

Pers	onnel Costs:		GS/Bange/	Months	Monthly		Proposed
PM	Name	Position Description	Sten	Budgeted	Costs	Overtime	FFY 1996
	David Evans	Biometrician I	17E	5.0	5,198	0 101 0	26.0
	PCN 115062	FBIII	18A	3.0	5.371	0 0	16.1
		FTII(Valdez)	9A	3.5	2,159	4.922	12.5
	-	FTII(Anchorage)	9A	2.0	1.945	1.800	5.7
		FTII(Kodiak)	9A	1.0	2,120	1,580	3.7
		FTII(Kenai)	9A	2.0	1,945	1,800	5.7
		FTIII(Cordova)	9A	2.0	2,439	3,593	8.5
	Seawan Gehlbach	FBI	14B	7.0	4,291	0	30.0
+	PCN 117021	FB-III	18L	1.0	6,333	0	6.3
							0.0
1							0.0
							0.0
		Subtota		26.5	31,801	13,695	
Tho	se costs associated with prog	ram management should be indicated by place	ment of an <b>*</b> .		F	Personnel Total	\$114.5
Trav	vel Costs:		Ticket	Round	Total	Daily	Proposed
PM	Description		Price	Trips	Days	Per Diem	FFY 1996
*	Fishery Biologist III and II tra-	vel to Anchorage for workshops	224	3	9	150	2.0
	Supervisory trips to Whittier		224	4	8	150	2.1
li -	Supervisory trips to Anchora	ge	224	4	8	150	2.1
	Supervisory trips to Seward		224	3	6	150	1.6
	Supervisory trips to Kodiak		610	3	5	150	2.6
	Supervisory trips to Kenai		300	4	7	150	2.3
							0.0
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Those costs associated with program management should be indicated by placement of an *. Travel Total						\$12.7	
<b></b>				- " ,			
							FORM 3B
	1000					Personnel	
1996 Project Title: Coded Wire Tag Recoveries from Pink Salmon, PWS					& Travel		
		Agency: AK Dept. of Fish & Game					
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## 1996 EXXON VALDEZ TEUSTEE COUNCIL PROJECT BUDGET October 1, 1995 - September 30, 1996

Contractual Costs:				Proposed
Description				FFY 1996
lag Lab Costs				75.0
Air Charters for brood stock samp	ling			2.0
Air Charters for Supervision and D	Pata Transport			12.2
Dept. of Transportation Vehicle Re	ental		Ì	3.4
Office Costs	,			3.6
Renting Magnetic Tag Detectors				4.4
		١		
When a non-trustee organization i	s used, the form 4A is required.	Contractual	Total	\$100.6
Commodities Costs:				Proposed
Description				FFY 1996
		Commodities 1	Total	\$2.9
1996	Project Number: 96186 Project Title: Coded Wire Tag Recoveries from Pink Salmon, PWS Agency: AK Dept. of Fish & Game		FO Cont Com D	RM 3B ractual & modities ETAIL
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# 1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET October 1, 1995 - September 30, 1996

New Equipment Purchases:		Number	Unit	Proposed
Description		of Units	Price	FFY 1996
				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 r	replacement equipment should be indicated by placement of an R.	New E	uipment Total	\$0.0
Existing Equipment Usage:			Number	Inventory
Description			of Units	Agency
				s.
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	Project Number: 96186			FORM 3B
1996	Project Title: Coded Wire Tag Recoveries from Pink Salmon. P	ws		Equipment
	Agency: AK Dent of Fish & Game			DETAIL
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#### Otolith Thermal Mass Marking of Hatchery Reared Pink Salmon in Prince William Sound

Project Number:	96188 (was 95320C)
Restoration Category:	General Restoration
Proposer:	ADF&G
Lead Trustee Agency:	ADF&G
Cooperating agencies:	Prince William Sound Aquaculture Corporation
Association	valuez risheries bevelopment
Duration:	Four years
Cost FY 96:	\$95,200
Cost FY 97:	\$100,500
Cost FY 98:	\$100,500
Cost FY 99:	\$48,800 (Closeout)
Cost FY 00:	\$0
Cost FY 01:	\$0
Cost FY 02:	\$0 .
Geographic Area:	Prince William Sound
Injured Resource/Service:	Pink Salmon

#### ABSTRACT

This project will develop otolith mass marking as an inseason stock separation tool for pink salmon in Prince William Sound. Inseason stock composition data is used by fishery managers to protect damaged wild pink salmon stocks from overharvest in mixed-stock fisheries. Coded-wire tags are presently used for this purpose in the Sound. Otoliths are small bones in the inner ear of fish. These bones can be marked through systematic changes in water temperature during egg incubation. The resulting marks are bands of light and dark material in the otolith similar to the bands in a tree. These induced marks can be used to identify hatchery-produced salmon in mixed-stock fisheries. Because all hatchery-produced salmon are marked using this technique, the cost of catch sampling is expected to be reduced, and the precision of inseason stock composition estimates is expected to be improved. The increased precision of stock composition estimates will improve the fishery manager's ability to protect damaged wild pink salmon stocks in mixed-stock fisheries.

This project will be conducted cooperatively by the Alaska Department of Fish and Game, Prince William Sound Aquaculture Corporation (PWSAC), and Valdez Fisheries Development Association (VFDA). In 1995, PWSAC and VFDA installed the necessary equipment to otolith mark all pink salmon embryos in the Armin F. Koernig, Wally H. Noerenberg, Cannery Creek, and Solomon Gulch hatcheries. Otolith thermal marking will begin after the embryos have passed the eyed stage of development in October 1995. Heated water will be introduced at the hatchery head troughs allowing treatment of millions of pink salmon embryos simultaneously.

The project will be conducted over two pink salmon lifecycles, marking both odd- and even-broodline fish. Experience with two complete lifecycles is needed to fully develop a program that integrates induced banding code quality, otolith processing rates and costs, and statistical designs for catch sampling. The feasibility and costeffectiveness of sampling the commercial catch for otoliths will depend upon whether a representative sample can be collected from the fishery. Several sampling techniques will be evaluated in 1996 using fin-clip experiments to determine if a truly random sample is obtained from each tender load of fish.

When otolith marked fish return as adults in 1997 and 1998, approximately 13,000 pink salmon otoliths will be processed in each year to estimate stock composition and corresponding confidence levels in PWS fisheries. The catch sampling program will also evaluate the variation in stock composition among tenders as well as between processors. A cost function for catch sampling will also be developed to produce an optimum allocation of sampling resources among tenders and processors. Monte Carlo simulation techniques will be used in conjunction with the data collected in this study to assess sampling power and refine sample sizes. It is anticipated that this project will overlap with coded-wire tagging for two years to allow for thorough evaluation of the otolith technique. This overlap period will also enable fishery managers to refine wild and hatchery pink salmon harvest estimates obtained using the coded-wire tag method.

#### INTRODUCTION

Each year approximately one half billion wild pink salmon fry emerge from the streams of Prince William Sound (PWS) and migrate seaward. Adult returns of wild pink salmon to PWS averaged approximately 10 million fish annually over the last two decades. The huge fry outmigrations and subsequent adult returns of pink salmon play major roles in the PWS 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 PWS because of their contribution to commercial, sport, and subsistence fisheries in the area.

Up to 75% of pink salmon spawning in PWS occurs in intertidal areas. In the spring of 1989 oil from the T/V Exxon Valdez oil spill (EVOS) was deposited in layers of varying thickness in intertidal portions of many western PWS streams utilized by spawning salmon. Pink salmon eggs 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 comparable and nearby unoiled streams in 1989, 1990, 1991, and 1992. Weidmer (1992) also observed a high incidence of deformities and elevated levels of cytochrome P-450 among fry in oiled streams in 1989. Willette (1993) reported reduced growth and survival of pink salmon fry and juveniles which reared in oiled marine waters of PWS 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.

PWS pink salmon returns originating from brood years subsequent to the EVOS have been aberrant or weak. Returns of wild and hatchery pink salmon in 1991 were only slightly below the mid-point of the preseason forecast but arrived late and had very compressed run timing. The fish were also small and in advanced stages of sexual maturity long before reaching their natal streams. As a result of this small size and advanced maturity, the fish 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 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.

Although hatchery pink salmon production in PWS began in the 1970's, returns from maximum permitted levels of fry production did not occur until the late 1980's and early 1990's and coincided with the EVOS Wild salmon populations injured by the EVOS are exploited in era. mixed stock commercial, sport, and subsistence fisheries which are dominated by returns from more productive hatchery populations. Wild pink salmon populations originate from hundreds of streams in PWS. Migratory timing and abundance of wild returns in marine fishing areas varies among populations. To sustain production from wild populations managers must insure that adequate numbers of wild fish from all portions of the wild return 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 this end, managers must be able to distinguish wild from hatchery fish and estimate their relative spatial and temporal abundance in fishing areas.

In addition to their dominance in the catch, hatchery stocks may also complicate management of PWS fisheries by straying into streams and spawning with wild fish. The magnitude and range of straying by both hatchery and wild pink salmon stocks in PWS may significantly
influence the success or failure of restoration efforts directed at wild stocks. The definition of what constitutes a wild population and the scale of restoration efforts may change if significant straying also occurs among wild populations. If straying of hatchery fish is significant and does lower the fitness of wild populations, restoration efforts which concentrate on insuring that spawning escapement goals are met may fail if no attention is given to the origins of the escapement.

#### 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 PWS. The methodology has been used extensively to estimate hatchery and wild stock contributions to commercial harvests and has also been used in preliminary straying research. Despite its usefulness, there are drawbacks 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 also intrusive, tags can be shed, and tagging may affect subsequent survival. Tag loss through shedding and differential mortality of tagged individuals affects subsequent estimates of adult returns based on tag recoveries. There is also recent evidence that poor placement of coded-wire tags may cause salmon to stray.

#### B. Rationale

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This project will develop otolith mass marking as an inseason stock separation tool for salmon. Because of the cost and problems associated with coded-wire technology, other alternatives for marking larger portions of populations with relatively inexpensive nonintrusive methods must be investigated. By marking most or all of the fish in a population, sample sizes at the time of tag recovery may be much smaller without affecting the accuracy and precision of contribution estimates. Non-intrusive marks which cannot be shed and which do not affect survival or behavior will also eliminate important sources of error in mark-recapture population and straying rate This data is essential information used by fishery estimates. managers to reduce fishery exploitation rates on damaged wild salmon Coded-wire tags are presently used for this purpose, but stocks. otolith marking is expected to provide more accurate information at a Numerous studies have documented the induction of rings lower cost. of light and dark material on fish otoliths by manipulation of water temperature during embryonic stages (Bergstedt et al. 1990, Brothers Each of these E.B. 1990, Munk and Smoker 1990, Volk et al. 1990). 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 PWS, the Alaska Department of

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Fish and Game (ADF&G) and Prince William Sound Aquaculture Corporation (PWSAC) reviewed the feasibility of otolith thermal marking at PWS hatcheries as well as otolith recovery in the commercial fisheries (Geiger et al. 1994). This review identified development of an inseason otolith sampling and mass processing program as the area where additional work was needed to fully develop otolith thermal marking technology for application as a large-scale inseason commercial fisheries management tool.

#### C. Summary of Major Hypotheses and Objectives

The rationale for this project is based on the hypothesis that otolith mass marking will provide a more cost effective and precise method for identification of wild and hatchery pink salmon in PWS. The principal objective of the project is to develop a large-scale inseason stock separation program using otolith thermal marks for use by fishery managers. The increased precision in stock composition estimates by time and area is expected to increase the manager's ability to protect damaged wild salmon stocks in mixed-stock fisheries.

#### D. Completion Date

All objectives of this multi-year project are expected to be met by FY 98. At that time, support for a fully developed inseason stock separation program will likely be shared by the ADF&G and the private sector.

#### COMMUNITY INVOLVEMENT

This project was developed through three months of ecosystem research planning by the Prince William Sound Fisheries Ecosystem Research Planning Group (PWSFERPG) as part of the Sound Ecosystem Assessment (SEA) program. The PWSFERPG conducted public meetings each week in the fall of 1993. Scientists from the University of Alaska, University of Maryland, Prince William Sound Science Center, PWSAC, ADF&G, and U.S. 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 SEA.

This project is partially sponsored by the PWSAC, the regional private, non-profit (PNP) aquaculture association for PWS, and Valdez Fisheries Development Association, a small PNP association. Development of mass marking programs, such as the PWS coded wire tagging program, has been a cooperative effort between ADF&G and PWS 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, review coded-wire tag project plans and results annually before approving subsequent funding. Operational plans and results of mass marking projects are also reviewed periodically by the PWS/CR Regional Planning Team as well as interested fishing industry groups. As part of the Trustee Council NRDA and Restoration process the code-wire tag mass 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 PWSAC board of directors in 1993 and, the Alaska Board of Fisheries in 1994. The PWSAC and VFDA board of directors as well as the PWS/CR Regional Planning Team have endorsed development of otolith thermal mass marking of hatchery salmon in PWS.

#### FY 96 BUDGET

Pers	onnel	59.2
Trav	el	2.4
Cont	ractual	16.9
Supp	lies	1.6
Equi	pment	5.0
-	Subtotal	85.1
Gen.	Admin.	10.1
	Total	95.2

#### PROJECT DESIGN

#### A. Objectives

A total of five major objectives will be achieved when the project is completed. Two of these will be achieved during FY 96 and all subsequent fiscal years:

- 1. Apply otolith thermal marks to all pink salmon embryos rearing in the AFK, WHN, CCH, and SGH hatcheries.
- 2. Evaluate the quality of otolith thermal marks applied to pink salmon embryos at AFK, WHN, CCH, and SGH hatcheries and collect voucher samples.

The three remaining objectives will be achieved in FY 97 and 98:

- 3. Evaluate methodology for collecting random samples from tender boats.
- 4. Estimate stock composition of commercial catches of pink salmon using otolith thermal marks.
- 5. Determine optimal allocation of sampling effort and estimate sample sizes.

#### B. Methods

#### **Objective** 1

Pink salmon will be marked during the eyed egg to hatch stage at PWS hatcheries. This approach will eliminate the need to degas the

incubation water. Gas saturation is usually not a problem for salmon embryos prior to hatch. Salmon eggs maintain a positive internal pressure which allows them to tolerate total dissolved gases (TDG) up to 110-116%. It would be uncommon to have TDGs of greater than 110% in incubation process water, but it may be possible to drive TDGs this high through aggressive heating. TDGs will be monitored during the thermal marking process. After hatch, gas supersaturation may cause salmon alevins to develop gas bubble disease. Expensive degassing equipment would be required to otolith mark pink salmon alevins.

A unique otolith thermal banding code will be used for each pink salmon hatchery in PWS. A unique hatchery mark will provide consistency in both application and recovery of the mark. The thermal mark will be applied in the eyed-egg to hatch zone of the otolith. The eyed-egg to hatch window occurs between October and December with an average length of 35 days. Approximately 22 days will be required to apply the thermal banding code at each hatchery. The hatcheryspecific codes will be composed of 5-7 thermal rings (Table 1). A single code for each hatchery will allow estimation of survival rate by hatchery. However, hatchery operators may also need to estimate survival rate for three treatment groups within each hatchery. In this case, a treatment-group code composed of three thermal rings will be applied in addition to the hatchery-specific basemark to distinguish among treatment groups.

Table 1. Proposed basemarks for PWS pink salmon hatcheries. The thermal schedule describes the actual temperature regime. The letter "H" refers to relatively hot water, while "C" refers to relatively cold water; the difference between the two temperature levels being 3.5 degrees Centigrade. The number directly before the thermal level is the number of rearing-hours at that level. Numbers in parenthesis before an "X" denote the number of repetitions.

Facility Pattern		Thermal	Schedule	Banding	
IIII	Cannery Creek	(3X)48H:24C,(	(1X)96H:24C,(3X)48H:24C	III	
III	WHN	(4X)48H:24C,	(1X)96H:24C, (2X)48H:24C	IIII	
	AFK	(5X)48H:24C		IIIII	
	VFDA	(7X)48H:24C		IIIIII	

#### **Objective 2**

Quality control during mark application is an important part of the otolith thermal marking program. Quality control is related to mark decoding, since it will largely determine a reader's ability to properly identify the mark. The placement of the thermal banding code on the otolith is critical to mark quality. The banding code will be applied by lot (i.e. a group of eggs taken on a single day) or groups of lots, when embryos are at the appropriate stage of development. Each incubating appliance will be sampled to ensure the mark was correctly applied. We expect that developmental stage and thus basemark placement will differ among lots within the hatchery. Temperature recorders will be installed at various points in the incubation system during mark application to document temperature changes.

A stratified-random sampling design will be used to estimate the proportion of unmarked otoliths at each PWS pink salmon hatchery (Cochran 1977). One month after mark application, a random sample of alevins will be taken from each lot, preserved in 100% ethanol, and sent to the ADF&G Otolith Laboratory in Juneau. Sample sizes will be selected in proportion to lot size, but a minimum of 100 alevins will be taken from each lot. At least thirty alevins will also be collected from each of 20 streams during the annual pre-emergent fry survey conducted by ADF&G. The samples will be used initially to validate that each hatchery-specific code was properly applied. Blind tests will then be conducted to estimate the proportion of alevins. marked at each hatchery. A reader's ability to distinguish hatcheryspecific codes, and marked otoliths among unmarked otoliths will used to determine the proportion marked. The set of otoliths for the blind tests will be obtained from a random subsample of alevins (n=300)taken from each hatchery sample combined with 600 wild alevins (total Samples from all sources will be randomly combined to 1800 otoliths). construct six test sets of otoliths (n=300). This test design will result in a composition of otolith types very similar to that encountered in samples taken from the commercial fishery when the fish return as adults. Two blind tests will be conducted with each of three readers.

Blind tests will be conducted at the ADF&G Otolith Laboratory in Juneau. After the otoliths are extracted from the alevins, they will be fixed to a glass slide with thermo-plastic cement. A grinding wheel will be used to remove material from one side of the otolith and expose the internal structures. The depth of grinding will be monitored by repeated viewing under a dissecting microscope. After the internal bands are exposed, the thermal mark will be decoded under a compound microscope.

#### <u>Objective 3</u>

The feasibility and cost effectiveness of sampling the commercial catch for otoliths will depend upon obtaining a truly random sample from the harvest. This project component will focus on development of a methodology for collecting random samples from tender boats unloading salmon onto conveyor belts at processing plants. Twenty

experiments will be performed during the 1997 fishery (FY 97) to determine whether random samples can be obtained. During each experiment, ADF&G technicians will (1) monitor the total number of salmon loaded on a tender, and (2) add 1,000 externally marked salmon (e.g. with clipped dorsal fins) to the load. The placement of marked salmon in the load would be varied for different experiments. sample of 510 salmon from each of these 20 tenders will be collected as salmon are unloaded and moved along conveyor belts into processing plants. If random samples are obtained, the estimate of the proportion of marked salmon in each tender load will fall within plus or minus 5% of the true proportion 19 out of 20 times. If our sampling design fails to meet this criterion, modifications will be made and further tests performed in 1997 and 1998. A more detailed description of otolith catch sampling experiments will be presented in the FY 97 detailed project description.

#### <u>Objective 4</u>

The stock composition of pink salmon catches will be estimated using otolith thermal marks in 1997 and 1998. Technicians will sample all tender boats delivering PWS pink salmon to processors in southcentral Alaska. The catch sampling program for recovery of otolith thermal marks will be highly integrated with the existing coded-wire tag recovery program. A considerable cost savings will be realized as a result. Technicians will employ a tender boat sampling methodology developed during the 1996 season. Inseason mass processing of otoliths will be conducted at the ADF&G otolith laboratory in Juneau. A more detailed description of the otolith catch sampling and mass processing program will be presented in the FY 97 detailed project description.

#### <u>Objective 5</u>

The proposed catch sampling program will estimate (1) the variance around estimates of the number of otolith-marked salmon within and among tender boats and among processors, and (2) the cost function for the otolith sampling program. This information will be used to estimate sample sizes and to develop an optimum allocation of sampling resources among tenders and processors (Cochran 1963). A more detailed description of this component of the project will be presented in the FY 97 detailed project description.

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  - C. Contracts and Other Agency Assistance

The ADF&G Commercial Fisheries Management and Development Division will ensure (1) that information obtained from this project is adequately documented and catalogued, and (2) that biometrics review of project methods and data analyses is obtained. The ADF&G Otolith Laboratory will process all otolith samples collected during this project. Since this project is a cooperative study conducted jointly by the ADF&G, PWSAC, and VFDA, contractual service agreements will be needed for application of thermal marks at each hatchery.

#### D. Location

This project will be conducted in the PWS region. Embryos will be thermally marked at the AFK, WHN, CCH, and SGH hatcheries operated by PWSAC and VFDA. Otolith code development and quality control work will be conducted at the ADF&G Otolith Laboratory in Juneau. In future years, an otolith catch sampling program will be developed. Catch sampling will likely occur in all PWS communities (e.g. Cordova, Valdez, Whittier), as well as, in Anchorage, Kenai, and Kodiak. Data analyses and reporting will be completed by ADF&G staff in Cordova and Anchorage.

#### SCHEDULE

#### A. Measurable Project Tasks for FY 96

This project will be conducted over one pink salmon life cycle for both the odd- and even-broodline populations. Embryos will be otolith marked in the fall of 1995 and 1996. Salmon from the 1995 and 1996 year classes will return to PWS as adults in the summers of 1997 and 1999. The following tasks will be accomplished in FY 96:

Start-up to December:	Apply thermal marks to BY 95 embryos at four nink salmon batcheries
January - February:	Collect samples from incubators to evaluate
	thermal mark quality
March - June:	Process and evaluate otoliths
April 15:	Submit annual project report for FY 1995
July - September:	Analyze data, make recommendations, develop FY 97 DPD
April 1997:	Submit annual project report for FY 1997

#### B. Project Milestones and Endpoints

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

December 1995:	Objective 1 - Apply thermal marks to brood year 1995 embryos
December 1996:	Objective 1 - Apply thermal marks to brood year 1996 embryos
June 1996:	Objective 2 - Evaluate thermal mark quality for brood year 1995
June 1997:	Objective 2 - Evaluate thermal mark quality for brood year 1996
September 1997:	Objective 3 - Evaluate tender sampling methodology
February 1998:	Objective 4 - Estimate harvest stock composition for brood year 1995
February 1999:	Objective 5 - Estimate harvest stock composition for brood year 1996

#### C. Project Reports

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

#### COORDINATION AND INTEGRATION OF RESEARCH EFFORT

The Otolith Mass Marking Project (96320C) is integrated with several other salmon restoration projects in PWS. This project will complement the Sound Ecosystem Assessment (SEA) program (Project 96320). SEA is a multi-disciplinary program designed to develop an understanding of the mechanisms regulating ecosystem function in PWS. SEA is focused on interactions of pink salmon and herring with other components of the PWS 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 SEA 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) develop a tagging program to estimate juvenile salmon mortality within PWS and the Gulf of Alaska. The Salmon Predation component of SEA will utilize otolith marked juvenile salmon to determine if predators select wild or hatchery salmon.

Project 94192, Hatchery Salmon Straying, was deferred to 1997 to allow development of otolith thermal marking technologies in PWS. Without the availability of a non-intrusive mass marking methodology it is unlikely that reliable estimates of total return, survival, and straying rates for wild salmon populations can be obtained. Therefore, the monitoring, research and restoration objectives of this project are also important to the Pink Salmon Genetics and the Pink Salmon Egg and Alevin Mortality projects.

The existing ADF&G fishery management program in PWS will provide salmon catch data needed to complete this project. An ADF&G preemergent fry program would provide otolith samples from wild salmon stocks in PWS. However, this program is not in the agency base budget and may not be operated in 1996. The ADF&G permanent staff of biologists and biometricians will write operational plans and provide overall supervision for this project. PWSAC and VFDA will use thermal mass marking to place unique marks on the otoliths of all pink salmon fry released from their facilities beginning in brood year 1995. The ADF&G Otolith Laboratory in Juneau will process all otoliths recovered from experiments and recovery operations.

#### ENVIRONMENTAL COMPLIANCE

This project has qualified for a categorical exclusion to the requirements of the National Environmental Policy Act.

#### 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

EMPLOYMENT: 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. Conducted 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. Conducted fisheries oceanographic studies in PWS in cooperation with private hatcheries and University of Alaska investigators. Chairman of PWS Regional Planning Team. Principal Investigator for Natural Resource Damage Assessment Study FS4A: Injury Assessment for Juvenile Salmon in Prince William Sound; Restoration Project R105: Survey and Evaluation of Instream Habitat and Stock Restoration Techniques for Wild Salmon in Prince William Sound; Restoration Project 93024: Restoration of the Coghill Lake Sockeye Salmon Stock.

March 1986 - February 1991: Fisheries Instructor/ Assistant Research Professor, University of Alaska Fairbanks, School of Fisheries & Ocean Sciences, supervised by Dr. Don Kramer. Conducted research on effects of oceanographic conditions on 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, and salmon feeding on the high seas. Designed and implemented a program of education, research, and public service to promote fisheries development in northwest Alaska. Taught college level course in oceanography as well as marine safety and vocational training courses in fisheries.

#### EDUCATION:

- 1985 Master of Science, Fisheries Oceanography, University of Alaska Fairbanks.
- 1983 Bachelor of Science, Fisheries Science, University of Alaska Fairbanks.

PETER HAGEN - Cooperating Investigator (Otolith Laboratory Director) Department of Fish and Game, Commercial Fisheries Management and Development Division P.O. Box 20

#### Douglas Alaska, 99824-0020

#### EMPLOYMENT:

August 1991 - Present: Director of The Alaska Department of Fish and Game's 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 fish in mixed stock fisheries. The laboratory is also charged with aging groundfish using otoliths and other hard structures. The ageing 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 substocks 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 PO Box 240020 Douglas, AK 99824

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 ADF&G-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; ageing scales; collecting fish using various trapping methods. Hatchery and weir experience includes installing, operating, and maintaining weirs; collecting data and keeping records; tagging (CWT) 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 ageing 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 333 Raspberry Rd.

#### Anchorage, Alaska 99518

#### EMPLOYMENT:

October, 1991 - present: Biometrician I with Alaska Dept. 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:

Master of Science, Statistics, Oregon State University
Doctor of Philosophy, Soil Science, University of
Guelph (Ontario, Canada)
Master of Science, Soil Science, University of Guelph (Ontario, Canada)
Bechelor of Science, Soil Science, University of
Nottingham (U.K.)

<u>ن</u> :

Mark Willette, Interim Principal Investigator Alaska Department of Fish and Game Commercial Fisheries Management and Development Division P.O. Box 669 Cordova, Alaska 99574-669 (907) 424-3214 (phone) (907) 424-3235 (FAX) . Dull. Joseph/Sullivan, Fisheries Program Manager Alaska Department of Fish and Game Habitat and Restoration Division 333 Raspberry Rd. Anchorage, Alaska 99518-1599 (907) 267-2213 (phone) (907) 344-3148 (FAX)

Date prepared

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

<u> </u>	Authorized	Proposed						
Budget Category:	FFY 1995	FFY 1996						
Personnel	\$25.7	\$57.5						
Travel	\$1.4	\$2.4						
Contractual	\$577.4	\$16.9						
Commodities	\$10.4	\$1.6						
Equipment	\$8.2	\$5.0		LONG F	RANGE FUNDIN	IG REQUIREMEI	NTS	
Subtotal	\$623.1	\$83.4	Estimated	Estimated	Estimated	Estimated	Estimated	Estimated
General Administration	\$27.7	\$9.8	FFY 1997	FFY 1998	FFY 1999	FFY 2000	FFY 2001	FFY 2002
Project Total	\$650.8	\$93.2	\$100.5	\$100.5	\$48.8	\$0.0	\$0.0	\$0.0
Full-time Equivalents (FTE)	0.6	0.9						
			Dollar amoun	ts are shown in	thousands of a	dollars.		
Other Resources								
Comments:								
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	<u></u>	<u> </u>			<u></u>			
	[······							
[	Project Num	ber: 96188						FORM 3A
	Project Title	Otolith The	rmal Mass M	larking of Ha	tchery Pink	Salmon		AGENCY
1996						Jannon		
		in Prince	william Sour	10				
	Agency: Ak	C Dept. of Fi	sh & Game					DETAIL
Prepared: 1 of 2	<u></u>							7/19/05

1 of **2**4

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

Pers	onnel Costs:		un		GS/Range/	Months	Monthly		Proposed
PM	Name		Position Description		Step	Budgeted	Costs	Overtim	e FFY 1996
	Vacant		Fishery Biologist III		18D	3.0	5,915		0 17.7
	Kris Munk		Fishery Biologist I		14E	1.0	4,676		0 4.7
	Vacant		Fish & Wildlife Technician III		11A	3.0	3,509		0 10.5
	D. Evans		Biometrician		19C	3.0	6,095		0 18.3
*	PCN 117021		Fishery Biologist III		18L	1.0	6,333		0 6.3
									0.0
									0.0
									0.0
									0.0
									0.0
									0.0
<b> </b>			1	Subtatal		11.0	26 529		0.0
Tho	Subtotal Those costs associated with program management should be indicated by place					11.0	20,528	Personnel Tot	al \$57.5
Tray	Travel Costs:				Ticket	Round	Total	Dail	v Proposed
PM	Description				Price	Trips	Days	Per Dier	n FFY 1996
	1								0.0
	Cordova-Anch	. to attend ann	ual EVOS workshop		200	1	4	9	5 0.6
1	Anch Cordov	va for biometric	es support on catch sampling		200	2	6	9	5 1.0
	Juneau - Cordo	ova for training	of hatchery staff on mark appl.		300	1	5	9	5 0.8
									0.0
									0.0
									0.0
									0.0
			. f.						0.0
									0.0
									0.0
The	Those costs associated with program management should be indicated by placement of an *							Travel Tot	al \$2.4
		ated with plog		ted by place					
Project Number: 96199							Г	FORM 3B	
	Project Number: 90100				f Untobony P	ink Salman in			Personnel
	1996			warking 0	i natchery P	IIIK Jaimun II			& Traval
			Prince Willia	am Sound					
Agency: AK Dept. of Fish & Game									DEIAIL

7/18/95

# 1996 EXXON VALDEZ TRUS COUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

Contractual Costs: Description		······································	Proposed FFY 1996
Air charter for training a Fuel for otolith thermal Fuel for otolith thermal	at hatcheries (5 hrs. @ 275 per hr) mark application at Solomon Gulch Hatchery (5200 gals. @ \$1.0 per gal.) mark application at three PWSAC hatcheries (10300 gals. @ \$1.0 per gal.)		1.4 5.2 10.3
When a non-trustee organiza	tion is used, the form 4A is required	Contractual Tot	al \$16.9
Commodities Costs:			Proposed
Description			FFY 1996
Cell culture trays Acetate compression pla Grinding paper Glass slides - regular Glass slides - petrograph Slide boxes vials, storage alcohol Misc forceps and gloves	ate hic s		0.1 0.2 0.2 0.3 0.3 0.3 0.2 0.2
		Commodities Tot	al \$1.6
1996	Project Number: 96188 Project Title: Otolith Mass Marking of Hatchery Pink Salmon in Prince William Sound Agency: AK Dept. of Fish & Game		FORM 3B Contractual & Commodities DETAIL

### 1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

New	Equipment Purchases:		Number	Unit	Proposed
Des	ription	·	of Units	Price	FFY 1996
					0.0
	Dissecting microscope	e (Wild M3z, 1.0X apo objective)	1	5,000	5.0
					0.0
	· ·				0.0
					0.0
					0.0
					0.0
ſ					0.0
					0.0
					0.0
					0.0
				5	0.0
The	l	d with replacement equipment should be indicated by placement of an P	Now E	winmont Total	65.0
Evic	ting Equipment Uses	d with replacement equipment should be indicated by placement of an R.	INGW E	Aupment Total	\$5.0
Existing Equipment Usage:				of Units	Agency
003	Inption				Agency
	Boiler module (Temp.	inc. 21 deg. F and 200 gpm)		1	VFDA
	Boiler modules (Temp	inc. 21 deg. F and 200 gpm)		2	PWSAC
	Boiler module (Temp.	inc. 21 deg. F and 200 gpm)		- 1	ADFG
				•	
		;			
		Project Number: 96188			FORM 3B
	1006	Project Title: Otolith Mass Marking of Hatchery Pink Salmon	in	F	Fauinment
	1990	Prince William Sound			
		Agency: AK Dent of Fish & Geme			DETAIL
L		Agency. AN Dept. of Fish & Game		L	

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# Construction of a Linkage Map for the Pink Salmon Genome

Project Number: Restoration Category: Proposer:

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Lead Trustee Agency: Duration: Cost FY 96: Cost FY 97: Cost FY 98 - 00: Geographic Area: Injured Resource: 96.190 Research Fred W. Allendorf University of Montana ADFG Five years \$240,000 \$250,000 to be determined Prince William Sound Pink salmon

### ABSTRACT

We propose to construct a detailed genetic linkage map for pink salmon by analyzing the genetic transmission of several hundred DNA polymorphisms. The ability to genetically map the 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.

### INTRODUCTION

We propose to construct a genetic linkage map for the pink salmon genome. Such a map would 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 would 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.

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 important significance for ongoing work with pink salmon under the project Oil-Related Embryo Mortalities (Restoration Study 95191A). That project proposes to

identify germline mutations in pink salmon exposed to oil. As explained in the FY95 Detailed Project Description (95191A), genetic damage induced by oil may either be small changes in nucleotide sequence (microlesions) or large-scale changes in chromosome structure (macrolesions). Restoration Study 95191A proposes to screen pink salmon DNA in order to detect such lesions. A detailed genetic map for pink salmon would be invaluable for interpreting the results of Restoration Study 95191A 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 oddand 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).

### NEED FOR THE PROJECT

### A. Statement of Problem

Elevated embryo mortalities were detected in populations of pink salmon (*Oncorhynchus gorbuscha*) 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 the 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. The genetic map we propose to construct 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 would 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.
- 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 would 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 pallasi*). Comparative gene mapping has shown that the linkage groups in a wide variety of vertebrates have been conserved. If we find that certain loci in pink salmon 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. Summary of Major Hypotheses and 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 in pink salmon. We will use several types of different genetic markers. The primary type will be so-called random amplified polymorphic DNA's (RAPD's) using the polymerase chain reaction (PCR). Our goal is to map several hundred of these loci so that we have a detailed saturated linkage map. We will use these RAPD loci as a basis for mapping other DNA polymorphisms (e.g., microsatellite loci), as well as loci encoding protein polymorphisms (allozymes). This genetic map will allow testing of several hypotheses of Project 95191A related to identifying sites of genetic damage (lesions) induced by exposure to oil. The primary hypotheses are: (1) Genetic lesions have been induced by oil exposure; (2) These lesions are caused by point mutations (microlesions); (3) These lesions are caused by chromosomal breakage and deletions (macrolesions).

Secondary objectives of this proposed research are to develop a large number of genetic markers for estimation of straying rates, stock separation, and management of pink salmon and for evaluating the success and potential detrimental effects of supplementation programs. A genetic map will also allow us to test the hypothesis that marine survival has a genetic basis to it. We also have a variety of specific genetic hypotheses that we will test as explained in the more detailed Project Design.

### **D.** 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 will be 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 propose to use the Alaska SeaLife Center Research Facilities at Seward when they are available. Such a facility will greatly strengthen genetic investigations with pink salmon by allowing multigenerational studies. We cannot estimate budget costs after the first two years without knowing the cost structure of using the Alaska SeaLife facility.

#### **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 an professional educator in a university I am very committed to educational efforts. These will include informational meetings in the communities of Prince William Sound, including the Alaska SeaLife Center in Seward, and articles in the Trustee Council newsletter.

### **FY 96 BUDGET**

Personnel	130.0
Travel	4.5
Commodities	23.5
Equipment -	22.0
Subtotal	180.0
Indirect Costs	60.0
Total	240.0

### **PROJECT DESIGN**

#### LITERATURE CITED

- Allendorf, F. W. and G. H. Thorgaard. 1984. Tetraploidy and the evolution of salmonid fishes. Pages 1-53 in B. J. Turner, ed. Evolutionary Genetics of Fishes. Plenum Publishing Corp., New York.
- Biggs, E., T. Baker, M. McGurk, J. E. Hose and R. Kocan. 1991. Injury to Prince William Sound Herring. State/Federal Natural Resources Damage Assessment Draft Preliminary Status Report. Unpub. rep. Alaska Department of Fish and Game, Cordova, AK.
- Gharrett, A.J. and W.W. Smoker. 1991. Two generations of hybrids between even-year and odd-year pink salmon (Oncorhynchus gorbuscha): A test for outbreeding depression? Can. J. Fish. Aquat. Sci. 48:1744-1749.
- Longwell, A. C. 1977. A genetic look at fish eggs and oil. Oceanus 20(4):46-58.
- Malkin, D. 1994. Germline p53 mutations and heritable cancer. Annual Reviews in Genetics 28:443-465.
- Mironov, O. G. 1969. The development of some Black Sea fishes in seawater polluted by petroleum products. Probl. Ichthyol. 9(6):1136-1139
- Moles, A., M. M. Babcock and S. D. Rice. 1987. Effects of oil exposure on pink salmon (*Oncorhynchus gorbuscha*) alevins in a simulated intertidal environment. Marine Environment Research 21:49-58.
- Postlethwait, J.H. and 14 other authors. 1994. A genetic linkage map for the zebrafish. Science 264:699-703.

### SCHEDULE

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

### ENVIRONMENTAL COMPLIANCE

Our laboratory is regularly screened by the Environmental Health Department of the University of Montana for compliance with all federal, state, and local environmental laws and regulations.

# PERSONNEL

Project Leader: FRED W. ALLENDORF

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:

1975-1	1976	Lektor, Department of Genetics and Ecology, Aarhus			
1076.1	1070	Assistant Professor of Zoology University of Montana			
1970-1	1070	NATO Fellow Genetics Research Unit University of			
1970-1	. 979	Nottingham England			
1070 1	109/	Associate Professor of Zoology University of Montana			
1979-1	1004	Visiting Scientist Department of Capation Univ. of			
1963-1	1904	California, Davis			
1984-1	989	Professor of Zoology, University of Montana			
1989-1	990	Program Director, Population Biology and			
		Physiological Ecology, National Science Foundation (NSF)			
1992-1	993	Visiting Professor. University of Oregon			
1990-		Professor of Biology, University of Montana			
1993-		Director, Organismal Biology and Ecology Graduate			
		Program, University of Montana			
HONORS:	NAT	O/NSF Postdoctoral Fellowship, University of Nottingham, 1978-1979			
	Euro	pean Molecular Biology Organisation (EMBO), Fellowship, University of Stockholm, 1979			
	Disti	nguished 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			
-	Burli	ngton Northern Faculty Achievement Award for Research.			
-		University of Montana, May 1991			
	Elect	ed Member, AAAS Council (Biological Sciences Division)			
-	Euro Disti Burli Elect Burli Elect	<ul> <li>1978-1979</li> <li>pean Molecular Biology Organisation (EMBO), Fellowship, University of Stockholm, 1979</li> <li>nguished Scholar Award, University of Montana, June 1985</li> <li>ngton Northern Faculty Achievement Award for Research, University of Montana, June 1987</li> <li>red Fellow, American Association for the Advancement of Science (AAAS), February 1987</li> <li>ngton Northern Faculty Achievement Award for Research, University of Montana, May 1991</li> <li>red Member, AAAS Council (Biological Sciences Division)</li> </ul>			

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, Population Biology, 1983-1986, \$121,000 National Science Foundation, Faculty Research Opportunity Award, 1986, \$10,000 United States Department of Agriculture (USDA) Grant, Aquaculture, 1983-1985, \$43,000 National Science Foundation Research Grant, Population Biology, 1986-1989, \$148,000 National Science Foundation, Dissertation Research Grant, Steve Forbes, 1988-1990, \$9,850 National Science Foundation Research Grant, Population Biology, 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-)

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,	National	Science	Foundation
	(1987-19	989)							

Panel Member, International Program, National Science Foundation (1987)

Panel Member, Conservation and Restoration Biology, National Science Foundation (1991-1992)

- 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, National Science Foundation (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

### FOREIGN POSITIONS/GRANTS:

1975-1976	Postdoctoral research position, Department of Genetics and Ecology, Aarhus University, Aarhus, Denmark.
Oct 1975	Visiting scientist, Department of Genetics, Stockholm University, Stockholm, Sweden.
1977	Participating scientist, Biochemical investigations of the evolution of genes and enzymes (C. Markert, Yale University, P. I.). Aboard Research Vessel Alpha Helix in Caribbean Sea (Belize, Honduras, and Panama). Nov-Dec.
1978-1979	NATO/NSF Postdoctoral Fellow, Laboratory of Professor Bryan Clarke, Genetics Research Unit, University of Nottingham, England.
Sep 1978	Genetics Society of America Grant to attend XIV International Congress of Genetics, Moscow, USSR.
1979	EMBO Short-term Fellow, Department of Genetics, Stockholm University, Stockholm, Sweden. May-Jun.
Dec 1983	Genetics Society of America Grant to attend XV International Congress of Genetics, Delhi, India.
Jan 1989	USDA supported visit to Republic of China (Taiwan) to confer and advise on genetics and conservation of biodiversity.
Aug 1993 -	Selected to organize workshop on Conservation Biology and Genetics at the XVII International Congress of Genetics, Birmingham, England.

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Date Prepared

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Laboratory and Field Examination of Oil-Related Embryo Mortalities That Persist in Pink Salmon Populations in Prince William Sound

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Research and Monitoring
Alaska Department of Fish and Game
Alaska Department of Fish and Game
Washington State University National Marine Fisheries Service, Auke Bay Laboratory
Continue population monitoring until Pink Salmon in Prince William Sound have recovered.
\$474.6K
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Prince William Sound
Pink Salmon

## ABSTRACT

Elevated embryo mortalities were detected in populations of pink salmon inhabiting oiled streams following the March 1989 <u>Exxon Valdez</u> oil spill. These increased rates of mortality persisted annually 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 this putative genetic damage include physiological dysfunction of individuals and reduced reproductive capacity of wild pink salmon populations. The purpose of this project is to continue to monitor the recovery of pink salmon embryos in the field, to provide laboratory verification of the field results, and to verify and identify the occurrence of genetic damages. Results of these studies may provide the first evidence that the germline of fish exposed to chronic or acute sources of oil pollution can be damaged.

#### INTRODUCTION

Elevated embryo mortalities were detected in populations of pink salmon <u>Oncorhynchus gorbuscha</u> inhabiting oiled streams following the March 1989 <u>Exxon Valdez</u> oil spill (EVOS). These increased rates of mortality persisted annually 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 this putative genetic damage include physiological dysfunction of individuals and reduced reproductive capacity of wild pink salmon populations.

These effects would likely persist in populations of pink salmon for a longer duration than would be observed in other vertebrates because of the tetraploid nature of the salmonid genome. Salmonids evolved through a gene duplication event 25 million years ago (Allendorf and Thorgaard 1984). Pink salmon basically possess a duplicate set of chromosomes (tetraploid instead of diploid); although, some of the duplicates have been lost through subsequent evolutionary processes. However, the extra genes found for many loci would mask deleterious recessive alleles. The effects of these deleterious mutations would be uncovered in the homozygotes formed through the mating of heterozygotes in subsequent generations.

The purpose of this study is to continue to monitor the recovery of pink salmon embryos in the field and to provide laboratory verification of the field results presented by Sharr et al. (1994a, 1994b) and Bue et al. (in press). In this study we will (1) survey the same streams examined during the Natural Resource Damage Assessment (NRDA) process for pink salmon embryos in order to monitor recovery, (2) collect mortality data on pink salmon embryos produced from gametes taken from oil contaminated and uncontaminated streams in southwestern Prince William Sound (PWS) and incubated under identical conditions, and (3) test embryos and fry of oil-exposed ancestry for presence of genetic aberrations.

#### NEED FOR THE PROJECT

#### A. Statement of the Problem

Pink salmon embryos and fry that incubated in the oiled intertidal spawning areas in Prince William Sound in 1989, 1990, 1991, 1992, and 1993 appear to have been adversely affected by EVOS. Oil was deposited in layers of varying thickness in the intertidal portions of streams utilized by spawning pink salmon during the spring of 1989. Pink salmon eggs deposited in 1988 (1988 brood year) emerged as fry through the oiled spawning gravel during the spring of 1989 and began feeding on oiled plankton. These fish showed decreased growth due to oiling (Willette and Carpenter 1993). 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. (in press) 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. in press). 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. (in press) found increased embryo mortalities in oiled streams during the 1991 fall Furthermore, significant differences in embryo mortality survey. 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 from 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 genetic damage acquired during embryonic development 1989. Elevated embryo mortalities at all tidal zones in oiled streams were again detected during the 1992 survey (Sharr et al. 1994b; Bue et al. in press). Hatchery incubation experiments 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).

In previous Restoration Projects (94191, 93003, R60C) we used flow cytometry to test for the presence of macrolesions in pink salmon embryos exposed to oil. Flow cytometry is a rapid analysis technique used to score the presence of macrolesions through detection of distortions in DNA content among populations of cells (McBee and Bickham 1988). Flow cytometry has become an established method for measuring the physical and chemical characteristics of cells and has been used to detect clastogenic effects of environmental toxicants in several species (McBee and Bickham 1988; Bickham 1990; Lamb et al. 1991), but we were unable to detect macrolesions in pink salmon exposed to oil using this method (Miller et al. 1994, Miller et al. in prep.).

In Restoration Project 94191 we contracted with Washington State University (WSU) for a pilot study to examine the use of androgenetic haploids to expose deleterious microlesions. Androgenetic individuals are obtained by enucleating eggs with gamma radiation before fertilization. The resulting progeny are haploid, containing only a single set of chromosomes from the male parent and none from the female. Pre-hatch mortality curves for these haploids are directly related to the presence and number of deleterious mutations (Armstrong and Fletcher, 1983). Advantages of this technique over more classical techniques include rapid early detection, ability to detect the effects of point mutations, and the ability to detect the presence of deleterious recessive alleles. The androgenesis technique is not widely used because of the requirement of a gamma radiation treatment. Initial results from the pilot study show that androgenetic haploids produced from sperm that has undergone low-dose irradiation to produce mutations do die at faster rates than haploids produced from nonirradiated sperm. The androgenesis screen will be extended to analyze pink salmon of known oiling history in Trustee Council Project 95191B and 96191B.

Additionally, mutational load will be measured in replicates of oiled and non-oiled control treatments from Project 95191B by using an array of polymerase chain reaction (PCR) -based DNA assays. Primer selection for PCR will focus upon three potentially useful categories of loci: (1) introns that have shown to be conserved among salmonid species, show some intraspecific variation, and for which we have substantial baseline information (e.g., introns C and D of <u>GH-1</u> and <u>GH-2</u>, Forbes et al. 1994; Linda Park, National Marine Fisheries Service, personal communication); (2) microsatellite loci that have high rates of natural mutation (Park and Moran 1994; Wright and Bentzen 1994); and (3) the hot spot regions (<u>HSR A-D</u>) that have been frequently associated with germline mutations in other species in the otherwise highly conserved tumor suppressor gene <u>p53</u> (Malkin 1994).

# B. Rationale

In this project we propose to: (1) continue monitoring embryo survival rates in oiled and reference streams, (2) repeat the hatchery incubation experiment for odd-year populations spawning in eight oiled and eight reference streams, and (3) conduct laboratory studies to screen samples for DNA lesions not detectable by flow cytometry. The successful pilot study conducted by WSU will be expanded to include androgenic examination of sperm collected from males of known oiling history during Restoration Project 95191B. We will conduct an inhouse screen for elevated rates of mutation at mutational hot spots (cf., Orita et al. 1989a, 1989b; Forbes et al. 1994), and we plan to incorporate the expertise of a consultant laboratory expert in techniques such as restriction endonuclease fingerprinting (REF; Liu and Sommer 1995), denaturing gradient gel electrophoresis (DGGE; Hovig et al. 1991; Brunel 1994), or other as appropriate to assist in the identification of loci at which mutations have taken place. Results from component 3 will be used to evaluate the 1989 through 1992 study results of Sharr et al. (1994a, 1994b) and Bue et al. (in press).

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 would provide the first evidence that the germline of fish exposed to chronic or acute sources of oil pollution can be compromised.

## C. Summary of Major Hypotheses and Objectives

In addition to monitoring pink salmon embryo mortalities at oiled and non-oiled study sites, in this project we will investigate the hypothesis that embryos exposed to oil in 1989 and 1990 accumulated deleterious mutations in the germline. We hypothesize that the elevate: embryo mortalities that persist in post-1990 year classes, in the absence of exposure to oil, may be the result of genetic damage acquired during embryonic development.

### D. 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 unimpacted 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 below.

# 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. The laboratory portion of the project will be moved to the Alaska Sealife Center in Seward when that facility is available. Again, local hire will be used when possible, and ADFG plans to participate in all of the educational and outreach programs scheduled for the Center.

#### FY 96 BUDGET

Personne	21	244.8
Travel		15.6
Contract	ual	136.2
Commodit	ies	31.2
Equipmer	nt	0.5
Sub	ototal	428.3
Gen. Adm	nin.	46.3
Tot	al	474.6

# PROJECT DESIGN

#### A. Objectives

The objectives of this project are to monitor the recovery of damaged pink salmon populations and to test the hypothesis that germline damage is responsible for the persistent embryo mortalities observed in streams that were oiled. Working objectives are:

- 1. Component 1. Recovery Monitoring of Injury to Pink Salmon Embryos in Prince William Sound.
  - a. Estimate the density, by tidal zone, of embryos in 31 streams using counts of live and dead embryos.
  - b. Estimate embryo mortality of pink salmon embryos in both oil contaminated streams and noncontaminated reference streams.
- 2. Component 2. Controlled incubation to evaluate the effect of physical stream characteristics.
  - a. Determine if the elevated mortalities of pink salmon embryos observed in oiled streams can be attributed to environmental factors.
- 3. Component 3. Laboratory examination of pink salmon gametes and embryos of crude-oil- exposed ancestry to assess genetic damage.

- a. Test for correlations between oil-exposed ancestry and mutations detected through DNA assays of selected introns, microsatellite loci, and mutational hot spot regions.
- b. Determine if elevated occurrence of deleterious recessive mutations can be detected in haploid androgens of oilexposed ancestry.
- 4. Combining Field Observations and Laboratory Results.
  - a. Determine if the elevated embryo mortalities observed in oiled streams in 1991 are explained by genetic damage to 1989 and 1990 embryos.

#### B. Methods

1. Recovery Monitoring of Injury to Pink Salmon Embryos in Prince William Sound

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 0.3 m², circular digs were systematically made along each transect using a high pressure hose to flush embryos from the gravel. Embrvos and fry were caught in a specially designed net.

The following data were collected for each tide zone transect during embryo sampling:

- (1)The sample date.
- The sample tide zone. (2)
- The start and stop time for each tide zone transect.
- (3) (4) Numbers and condition (live or dead) of embryos by species.
- A subjective estimate of the overall percent yolk sac (5) absorption for fry.

Data were transferred from field notebooks into a Lotus spreadsheet for editing and summarizing.

Pink salmon embryos were separated from chum O. keta and coho O. kisutch salmon embryos by their smaller size. Chum salmon embryos were separated from coho salmon embryos by their greater development and different coloration. An embryo was considered dead if it was opaque or discolored with coagulated lipids. Pink salmon fry were differentiated from chum salmon fry by their small size. Sampling often killed fry (especially newly hatched fry), so fry were only considered dead if decomposition was evident.

Data Analysis **b**.

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  $m^2$  (E_{ii}) will be estimated by:

$$\hat{E}_{ij} = \frac{\Sigma L E_{ijk}}{0.3 n_{ij}} , \qquad (1)$$

where  $LE_{ijk}$  is the number of live embryos found in the kth 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.

Fink salmon embryo mortality will be estimated for each stream using the following relationship:

(2) 
$$\hat{M}_{ij} = \frac{\sum (DE_{eijk} + DF_{eijk})}{\sum (LE_{eijk} + DE_{eijk} + LF_{eijk} + DF_{eijk})}$$

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 kth dig from stream i, zone j, collected during embryo dig e, respectively.

The Arcsin square root transformation will be examined as well as the

Logit transform of embryo mortality [ln (odds)].

$$Logit_{ij} = ln \left[ \frac{\Sigma (DE_{eijk} + DF_{eijk})}{\Sigma (LE_{eijk} + LF_{eijk})} \right]$$
(3)

Differences in embryo mortality will be examined using a mixed effects two-factor experiment with repeated measures on one factor (Neter et al. 1990):

$$Y_{ijk} = \mu_{...} + O_i + Z_j + (OZ)_{ij} + S_{k(i)} + e_{(ijk)}.$$
(4)

The two treatments will be extent of oiling,  $(O_i, 2 \text{ levels}; \text{ oiled and} non-oiled)$ , and height in the intertidal zone  $(Z_i, 4 \text{ levels}; 2.1, 2.7, and 3.4 m above mean low water, and upstream) both fixed effects. The data will be blocked by stream <math>(S_{k(i)})$ , a random effect nested within extent of oiling. The interaction of extent of oiling and height in the intertidal zone will also be examined. Equality of variances will be tested using the  $F_{max}$ -test (Sokal and Rohlf, 1981), while normality will be visually assessed using normal quantile-quantile and box plots (Chambers et al. 1983). If the data distribution appears to be nonnormal, 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). Different groupings of oiled and non-oiled streams will be analyzed if evidence of oiling is not consistent.

2. Controlled incubation to evaluate the effect of physical stream characteristics

a. Data Collection

In this component of the project we collect, analyze, and report the results of the 1995 brood year as well as initiate work on the 1996 brood year. We only intend to initiate new experimental matings during FY 96 if a difference is observed between embryo mortality in oil-impacted and unimpacted populations for the 1995 brood year.

This experiment will allow us to determine if results observed in NRDA Study F/S 2 and Restoration Studies R60C, 93003, and 94191 can be attributed to environmental factors. We will collect gametes from eight oiled and eight non-oiled reference streams from southwestern PWS, make intra-stream crosses, and incubate the resulting embryos in a controlled laboratory environment. Embryo mortality will be compared between the oiled and reference streams. If no difference is observed in this experiment, and if a significant difference in embryo mortality is detected between oiled and non-oiled streams during 1995 field sampling, then environmental factors probably account for the previous observations of elevated embryo mortalities.

Gamete collection and fertilization procedures will occur over a four day period to obtain data from eight oiled and eight non-oiled streams. Gametes from 30 male and 30 female pink salmon will be collected from two oiled and two control streams during each sampling day. The gametes will be flown to the Armin F. Koernig (AFK) hatchery where a random gamete pool will be assembled for each stream in a timely manner.

The random gamete pool will be constructed by placing approximately 30 eggs from each female (one teaspoon) into each of 30 cups. Each cup will then be fertilized by a different male. The 30 cups will be recombined into a large pail where the fertilized eggs will be mixed as they are rinsed. This method of creating a randomized gamete pool will insure that all possible crosses (30 x 30 = 900) will be present.

A minimum of nine randomly selected aliquots of approximately 500 embryos each will be collected from each intra-stream pool, placed into separate incubating vessels, and randomly placed into a common incubator.

Incubators will be periodically examined to count and remove dead embryos and score hatching success. The experiment will be terminated prior to the swimup stage at which time all larvae will be killed.

b. Data Analysis

The data will be analyzed as a fixed-effects randomized block design:

$$Y_{ijk} = \mu + B_i + O_j + e_{ijk}, \qquad (5)$$

where  $Y_{ijk}$  is embryo mortality for sample day i, oil contamination level j, and stream k;  $\mu$  is the model mean;  $B_i$  is sampling day a blocking variable;  $O_j$  is the level of oil contamination (oiled or not oiled); and  $\epsilon_{ijk}$  is random error. The relative power of the test was estimated (Neter et al. 1990), and the sample size was found sufficient to detect a difference of less than 1.5 standard deviations at  $\alpha$ =0.05 and 95% power. A test with high power is needed to protect against arriving at a false conclusion that the elevated embryo mortalities could be attributed to environmental factors when, in fact, they were not.

The assumption of constant error terms will be tested using the  $F_{max}$ -test (Sokal and Rohlf 1981) while normality will be visually assessed using scatter plots, box plots, and normal probability plots (Chambers et al. 1983). Appropriate transformations will be used to alleviate variance and normality concerns if they are detected. All suitable comparisons will be made using Bonferroni family confidence intervals. The SAS (SAS Institute Inc. 1988) General Linear Models Procedure will be used to analyze the data.

3. Laboratory examination of tissues from individuals of crude-oil exposed ancestry to assess genetic damage

In this component we will measure the genotoxic response of pink salmon to exposure to Prudhoe Bay crude oil. Controlled oiling was conducted over two brood years at the Little Port Walter field station by the National Marine Fisheries Service (Restoration Studies R60C and 93003). Mutational load will be measured in replicates of oiled and non-oiled control treatments using both an array of sensitive DNA assays and an androgenetic screen for deleterious recessive mutations. Sperm and tissues from adults subjected to oil as embryos, as well as their progeny, will be analyzed. This study will span two generations in order to evaluate the validity of the germline mutation hypothesis.

#### a. DNA Assays

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 buffered solution that protects the DNA from degradation; (2) a Proteinase K digest to deactivate the proteins; (3) an RNase treatment to digest RNA; (4) protein precipitation to remove Proteinase K, RNase, and denatured proteins; (5) isopropanol to precipitate the DNA; (6) 70% ethanol to wash the DNA; and finally (7) a hydration solution to rehydrate the DNA.

After extraction, the DNA will be amplified using the polymerase chain reaction (PCR; Saiki et al. 1988; Kocher et al. 1989; Chapman and Brown 1990; Carr and Marshall 1991). Primer selection for PCR will include loci from three potentially useful categories: (1) introns that are known to be conserved among salmonid species, show some intraspecific variation, and of which we have substantial baseline information (e.g., introns C and D of <u>GH-1</u> and <u>GH-2</u>, Forbes et al. 1994; Linda Park, National Marine Fisheries Service, personal communication); (2) microsatellite loci that have been shown to have high rates of natural mutation (Park and Moran 1994; Wright and Bentzen 1994); and (3) hot spot regions (<u>HSR A-D</u>) that have been most frequently associated with germline mutations in the otherwise highly conserved tumor suppressor gene <u>p53</u> in other species (Malkin 1994).

Genetic data will be collected using automated DNA assays. Fragment analysis for detection of restriction fragment length polymorphisms (RFLP) will be done following the methods of Forbes et al. (1994), except that data will be collected on an Applied Biosystems Incorporated (ABI) model 373 series automated sequencer. Sequence analysis, including SSCP screening (cf., Orita et al. 1989b), will be conducted on an ABI model 377 automated sequencer.

Additionally, a sister set of tissues will be provided to a consulting laboratory, obtained through the state procurement process, to a.d in the screening for genetic damage. Responses to a Request for Proposal (RFP) will be reviewed to select the best complimentary approach which may include alternative techniques such as REF, DGGE, heteroduplex analysis (Delwart et al. 1993), amplified fragment length polymorphism analysis (AFLP, Xue et al. 1993), or other approaches as identified through the peer-review process (cf., Brunel 1994; see rationale in Section d. Alternatives, below).

## b. Androgenesis

Androgenesis is a treatment in which eggs are treated with radiation before fertilization with normal sperm. If no other treatments are applied, the resulting offspring contain one chromosome set from the male and none from the female parent. Such haploid individuals survive until about the time of hatching and then die. If an additional heat or pressure treatment is applied to block the first cell division in the fertilized egg, diploid androgenetic offspring can be produced. These individuals can survive, although they tend to be weak because of inbreeding.

The relative survival of androgenetic haploids has been shown to be a sensitive measure of the presence of deleterious mutations carried by sperm from a given male (Armstrong and Fletcher 1983; Gary Thorgaard, Washington State University [WSU], unpublished data). The use of androgenetic haploids rather than androgenetic diploids is preferred because the diploids show poor pre-hatch survival due to the heat or pressure treatments. Additionally, androgenetic haploids are sensitive to recessive mutations that are lethal because both recessive and dominant mutations will kill haploid embryos, while only dominant mutations will kill normal embryos with one chromosome set from each parent. Recessive mutations are more likely to be the cause of the post-1991 embryo mortality, as dominant deleterious mutations would tend to be rapidly purged from the genome.

In this project, eggs and sperm from pink salmon will be collected in Alaska by ADFG, and the androgenesis will be conducted at Washington State University using the Cobalt-60 gamma source at the WSU Nuclear Radiation Center and WSU hatchery facilities.

Survival of androgenetic haploid individuals produced from 30 males from LPW oil-exposed treatments will be compared with the survival of androgenetic haploids produced from 30 males from LPW non-oiled controls. Each trial will be replicated three times. Using 100-200 eggs per replicate, about 20,000 unfertilized pink salmon eggs will be required. Use of the Cobalt-60 radiation source is the bottleneck of this experiment, so approximately 5,000 eggs will be shipped, with fresh sperm, to WSU at 4-5 day intervals to optimize application of the gamma-ray treatments. Sperm will be collected from individuals sampled for DNA assays (above), and results will be cross-referenced.

# c. Data Analysis

Genetic variation will be scored for the informative categories: introns, microsatellites, and <u>p53</u> HSRs. Individuals from both oilincubated and clean-incubated groups (up to 50 individuals from each treatment and control replicate available from the Little Port Walter experiments, see Restoration Science Project 94191B) will be examined using a randomized design for corresponding loci. Categorical data analysis will be used to test for differences in frequencies of genetic variants among treatments and controls.

ANOVA and survival analysis will be used to test for differences in mortalities obtained from the three replicates of treatment and control androgenic haploids.

# 4. Alternative Methodologies Considered

We conferred with individuals from some of the leading laboratories in the country working in this field and synthesized the input of three peer reviewers over three years prior to establishing the above protocol. Through this process we identified a number of procedures that are used to identify DNA damage in response to genotoxic challenge.

DNA adduct analysis developed into use as a molecular dosimeter of response to genotoxic compounds (reviewed in Reichart et al. 1994; see also Malins and Gunesman 1994). The correlation of sediment concentrations of mutagenic PAHs and hepatic tumors lead investigators to the understanding that the presence and persistence of PAH-DNA adducts are factors that directly relate to the carcinogenicity of a compound (Poirer et al. 1991; Reichart et al. 1994). Collier et al. (1994) found a correlation between PAH in the sediment and DNA adducts in oyster toadfish. However, germline mutations have not been indicated, and DNA adduct analysis was not recommended as a line of investigation to pursue (J. E. Stein and T. K. Collier, National Marine Fisheries Service, personal communication).

Several other short-term cytogenetic assays exist for evaluating the potential genotoxic effects of chemicals and compounds. These methods are designed to identify four general types of genetic changes: DNA microlesions, DNA macrolesions, primary DNA damage, and morphologic changes in target cells (Brusick 1987; however, some of the most promising approaches rely upon tissue culture techniques not yet successfully developed for salmonid tissues--R. M. Kocan, University of Washington, personal communication). Sister chromatid exchange (SCE) measurement has become a common technique for cytogenetic assays of primary DNA damage (Hsu 1982). The micronucleus test (MNT) and anaphase aberration (AA) counts have become standard measures of DNA macrolesions (Evans 1976; Kocan and Powell 1985; Kocan et al. 1985). These techniques are capable of detecting and quantifying subtle chromosome changes. However we identified limitations to these approaches for our purposes: (1) physical separation of metaphase and anaphase chromosomes for visual scoring is required; (2) techniques for chromosome separation and isolation can be technically involved and are not standardized between laboratories; (3) visual scoring of the desired endpoints can be subjective; and (4) time involved for isolating and scoring chromosomes limits sample sizes to 100-200 cells which reduces statistical accuracy and precision. Consequently, these cytogenetic approaches were not recommended for inclusion in this study.

Finally, flow cytometry has been demonstrated to be as sensitive as the AA test for detecting structural chromosome aberrations in dividing cells (Kocan and Powell 1985) and therefore provides a useful

technique for in vivo analysis of DNA macrolesions. Advantages of flow cytometry over other approaches are that it is less technically involved, easier to standardize, less time consumptive, and more statistically powerful. Flow cytometry can demonstrate the fate of chromosome/chromatid damage in subsequent generations of cells. For example, comparisons of  $G_1$  DNA content,  $G_1$  coefficient of variation, or presence of aneuploid cell populations can be used to test for the presence of chromosome damage (Cram and Lehman 1977; Bickham et al. 1988). Changes in the proportions of cells within the cell cycle may reflect a cytotoxic effect of a substance (Fertig and Miltenburger 1989). Flow cytometry allows analysis of large numbers of cells  $(10^3 -$ 10⁵) greatly increasing statistical power, a motivating force behind development of flow cytometry for cytogenetic testing (Deaven 1982). Sample preparation and measurement are reproducible, accurate, and can be completed in several minutes versus several hours for visual microscopic scoring (Otto and Oldiges 1980).

In Restoration Projects R60C, 93003, and 94191, we probed for macrolesions using flow cytometry. Useful results correlated exposures of very early embryos to seawater to the development of mosaic and triploid genomes. Further study documented that those genome aberrations were not responsible for the elevated embryo mortality observed in this series of studies (Miller et al. 1994). However, because flow cytometry was not sensitive enough to detect germline damage in the pink salmon embryos of known oiling history, we are redirecting our efforts to focus on more sensitive screens for microlesions (e.g., sequence-based analysis of mutational hot spots regions using an array of approaches, Ike Wirgin, NYU Medical Center of Environmental Medicine, personal communication). Further, previous reports of macrolesions detected through flow cytometry may be documenting genetic damage that is subject to DNA repair mechanisms and not persistent in the germline (cf., Liquori and Landolt 1985; R.M. Kocan, University of Washington, personal communication).

During our survey of the literature and contact with outside experts we identified that the technology for sensitive mutation screens is rapidly evolving. Modifications to the sensitive SSCP screen that we propose were released during the preparations of this proposal (Liu and Sommer 1995). A change in direction to focus upon a reversetranscriptase approach to the study of mutations present in mRNA at the time of embryo death was also suggested (Ike Wirgin, NYU Medical Center of Environmental Medicine, personal communication, February, Because of the quickly changing nature of this technology, 1995). ADFG scientists decided to reshape the study to include the support of postdoctoral researchers and/or applications specialists from outside sources, expert in technique development, to collaborate in the application of novel mutation screens. A number of university laboratories, the Applied Biosystems applications lab, and possibly the Environmental Conservation Division of the Northwest Fisheries Science Center, have expressed interest in collaboration.

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#### C. Contracts and Other Agency Assistance

The androgenesis subcomponent initiated by Dr. Gary Thorgaard, Washington State University (WSU), will be continued with WSU as a sole-source contractor. WSU is uniquely suited to conduct such a project. The WSU Nuclear Radiation Center has Cobalt-60 gamma radiation source that Dr. Thorgaard is currently using to conduct deleterious-mutation studies on rainbow trout. Dr. Thorgaard's laboratory is widely recognized as one of the leading laboratories in the world in the field of androgenesis in salmonids; to our knowledge it is the only laboratory in North America capable of such study.

We plan to replace the efforts of staff scientist Gary Miller, who has left ADFG, by supporting a post-doctoral position at one of the Alaskan universities through a Reimbursable Services Agreement.

Finally, based upon discussions with peer reviewers and other experts, we programmed \$50.5 K for a subcomponent to be awarded through the State of Alaska procurement process to provide for an applications laboratory to aid in the DNA assays using novel mutation screens. The cost was estimated based upon the current typical cost of funding a post-doctoral scientist at a university laboratory.

### D. Location

# Component 1:

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.

## Component 2:

Gametes for the controlled incubation to assess physical stream characteristics will be collected from as many as 16 streams in southwestern PWS - eight oiled and eight control. Embryo incubation will take place at the Armin F. Koernig hatchery in PWS.

## Component 3:

The exposure of gametes to oiled incubation substrate and their subsequent culture will be performed at the National Marine Fisheries Service Laboratory at Little Port Walter, Baranof Island, southeastern Alaska and are funded by Restoration Project 95191B. DNA sequencing will be done at the ADFG Genetics Laboratory in Anchorage. Androgenetic haploids will be produced and cultured at Washington State University. Additional DNA analysis will be done at a consultant laboratory to be determined by RFP.

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#### SCHEDULE

#### A. Measurable Project Tasks for FY 96

1.	Component 1. Recover Embryos	y Monitoring of Injury to Pink Salmon in Prince William Sound
15 30	Sep - 30 Oct 1995: Oct 1995 - 30 Mar 1996:	Embryo deposition sampling. Analysis of brood year 1995 embryo data and completion of first draft of 95191A
30	Oct 1996 - 30 Mar 1997:	Analysis of brood year 1996 embryo data and completion of first draft of 96191A report (97191A).
2.	Component 2. Controlled physical	d incubation to evaluate the effect of stream characteristics
1 7	Aug - 15 Aug 1995:	Preparation for brood year 1995 AFK
15	Aug - 30 Aug 1995:	Collect gametes and make crosses from 16 PWS streams; begin incubation of brood year 1995 gametes at AFK (95191A).
30	Aug - 15 Nov 1995:	Monitor incubators and collect data for brood year 1995.

15 Nov 95 - 30 Mar 96:	Analyze data for brood year 1995 and prepare first draft of 95191A report.
1 Aug - 15 Aug 1996:	Preparation for brood year 1996 AFK incubation experiment.
15 Aug - 30 Aug 1996:	Collect gametes and make crosses from 16 PWS streams; begin incubation of brood year 1996 gametes at AFK.
30 Aug - 15 Nov 1996:	Monitor incubators and collect data for brood year 1996 (97191A).
15 Nov 96 - 30 Mar 97:	Analyze data for brood year 1996 and prepare first draft of 96191A report (97191A).

3. Component 3. Laboratory examination of pink salmon gametes and embryos of oil-exposed ancestry to assess genetic damage*

1 Oct - 30 Oct 1995: Initiate haploid androgenesis and novel mutation screen contracts. 15 Aug - 30 Oct 1995: Obtain gametes, spawn second generation (one generation from oiling event). Send milt to University of Washington on contract to produce androgenetic haploids. Begin fertilized egg incubation. 15 Aug - 30 Oct 1995: Begin analysis of embryos at ADFG genetics laboratory. Continue fertilized egg incubation. 30 Oct 1995 - 15 May 1996: Continue analysis of tissues at ADFG genetics lab using mutation screens. 15 May - 30 Sep 1996: Evaluate ADFG results, subcontractor results, draft annual report

*All spawning, oiling, incubation, genetic sampling, and fish culture aspects will be done at Little Port Walter by the National Marine Fisheries Service under Restoration Project 95191B.

#### B. Project Milestones and Endpoints

Component 1.

Annual review terminate project component if embryo mortalities are not significantly different between oiled and nonoiled study sites for two consecutive years for both the odd- and two-even broodlines

Component 2:

Annual review terminate project component if results from laboratory studies of embryo mortality are the same as results from field study (e.g. component 1) Component 3:

Annual review terminate project component if results from aggregate of laboratory studies confirm no finding of genetic damage; modify project if warranted by findings of ADFG and collaborating laboratories; expand project to examine genetic damage in other species if findings warrant

## C. Project 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 96 will be submitted by March 30, 1997.

#### 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 follows most field activities of SEA (95320) and other pink salmon related projects 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 Cooperative Fisheries and Oceanographic Project, and these data will be utilized by several SEA studies.

Final edited data from all three components of this project will be stored electronically as computer databases, and final versions will be provided annually to the Information Modeling portion of SEA for incorporation into a centralized ecosystem database.

# ENVIRONMENTAL COMPLIANCE

Embryo sampling will require an ADFG Title 16 permit and an ADFG biological collections permit. An ADFG Fish Transport Permit will be required to obtain gametes from experimental streams and transport them to AFK hatchery for the controlled incubation component and to WSU for androgenesis studies.

## 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 and supervision of lab work, 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, ADF&G1991-Affiliate Associate Professor, U. of Alaska, Fairbanks1988-1990Assistant Professor, Southern Illinois University1987-1988Research Assistant Professor, University of Idaho1982-1986Graduate Research Assistant, University of Washington1980-1982Fish Biologist, Pacific Fisheries Research, Olympia,WA1978-1980Fish 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 (<u>Oncorhynchus keta</u>). J. Hered. 77:399-402.
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- Utter, F. M., and J. E. Seeb. 1990. Genetic marking of fishes: overview focusing on protein variation. Am. Fish. Soc. Sym. 7:426-438.
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- Seeb, J. E., and G. D. Miller. 1990. The integration of allozyme analyses and genomic manipulations for fish culture and management. <u>In</u>: D.H. Whitmore, Editor. Electrophoretic and Isoelectric Focusing Techniques in Fisheries Management. CRC Press, Boca Raton, pp 266-279.

- 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 <u>Salvelinus</u> species inferred from allozyme data. Can. J. Fish. Aquat. Sci. 51(Suppl. 1):182-197.

### B. Mark Willette, Area Research Biologist

Alaska Department of Fish and Game Commercial Fisheries Management and Development Division P.O. Box 669 Cordova, Alaska 99574 (907)424-3214

PROJECT RESPONSIBILITIES:	Design and supervision of embryo	
	surveys, analysis, report writing	J

EDUCATION: 1985 Master of Science, Fisheries Oceanography, University of Alaska Fairbanks 1983 Bachelor of Science, Fisheries Science, University of Alaska Fairbanks

**PROFESSIONAL EXPERIENCE:** 

1991		present	Area-Resource Development Biologist, Cordova,
			Commercial Fisheries Management and Developent, 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.

Christopher Habicht, Fisheries Biologist II C. Commercial Fisheries Management and Development Alaska Department of Fish and Game (907) 267-2385 Anchorage, Alaska 99518

**PROJECT RESPONSIBILITIES:** 

Supervision and conduct of hatchery matings, laboratory analysis, flow cytometric comparisons, coordination of laboratory and field efforts

# 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 <u>Exxon Valdez</u> 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.

D. Brian G. Bue, Biometrician II Commercial Fisheries Management and Development Alaska Department of Fish and Game Anchorage, Alaska 99518-1599 (907) 267-2123 BrianB%fishgame@state.ak.us

**PROJECT RESPONSIBILITIES:** Study design and analysis

EDUCATION: B.S., Fisheries, 1978, University of Alaska, Fairbanks B.S., Biology, 1978, University of Alaska, Fairbanks M.S., Fisheries, 1986, University of Alaska, Fairbanks

#### **PROFESSIONAL EXPERIENCE:**

1988- Biometrician II, CFMD, Alaska Dept. Fish and Game 1987-1988 Biometrician I, CFMD, Alaska Dept. Fish and Game 1978-1987 Fisheries Biologist I, CFMD, Alaska Dept. Fish and Game 1974-1977 Fish and Wildlife Technician, Alaska Dept. Fish and Game

SELECTED PUBLICATIONS AND PRESENTATIONS:

- Bue, B.G., S. Sharr, S.D. Moffitt, and A.K. Craig. <u>Ir Press.</u> Effects of the <u>Exxon Valdez</u> oil spill on pink salmon embryos and preemergent fry. <u>In Rice, S.D., R.B. Spies, D.A. Wolfe, and B.A.</u> Wright, editors. <u>Exxon Valdez</u> Oil Spill Symposium Proceedings. American Fisheries Society Symposium. <u>Accepted Pending</u> <u>Publication.</u>
- Geiger, H.J., B.G. Bue, S. Sharr, A.C. Wertheimer, and T.M. Willette. <u>In Press</u>. A life history approach to estimating damage to Prince William Sound Pink Salmon from the <u>Exxon Valdez</u> oil spill. <u>In</u> Rice, S.D., R.B. Spies, D.A. Wolfe, and B.A. Wright, editors._ <u>Exxon Valdez</u> Oil Spill Symposium Proceedings. American Fisheries Society Symposium. <u>Accepted Pending Publication</u>
- Rowell, K.A., H.J. Geiger, and B.G. Bue. 1990. Stock identification of Pacific Herring in the Eastern Bering Sea trawl bycatch and in the Dutch Harbor directed food and bait fishery. Proceedings of the International Herring Symposium, Alaska Sea Grant Report No. 91-01, Fairbanks.

- Bue, B.G. and D.M. Eggers. 1989. An age-length key for sockeye salmon smolt. Alaska Department of Fish and Game, Division of Commercial Fisheries, Regional Information Report No. 2D89-5, Anchorage.
- Bue, B.G. 1986. Effects of gill net selectivity on sockeye salmon in the Egegik and Naknek-Kvichak Districts, Bristol Bay, Alaska. Master's Thesis. University of Alaska, Fairbanks.
- Bue, B.G. 1986. Comparison of cable lay and center core gill net catches from the Port Moller offshore test fishery, 1985. Alaska Department of Fish and Game, Division of Commercial Fisheries, Bristol Bay Data Report No. 86-4, Anchorage.
- Bue, B.G., S. Sharr, G.D. Miller, and J.E. Seeb. 1995. Evidence of genetic damage in pink salmon inhabiting Prince William Sound, Alaska, three generations after the <u>Exxon Valdez</u> oil spill. Presented to the 1995 Pink and Chum Salmon Workshop, March, 1995, Bellingham, Washington.
- Bue, B.G. and R. Gates. 1991. Pink salmon escapement estimation using aerial observations. Presented to the Alaska Chapter of the American Statistical Association, September, 1991, Juneau.



Figure 1. Location of streams to be sampled for embryo deposition. 28

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Date Prepared

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1996 EXXON VALDEZ TRUS COUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

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Budget Category:     Authorized FFY 1996     Proposed FFY 1996       Personnel     \$176,9     \$38.3       Travel     \$22.3     \$4.2       Commodities     \$32.2     \$19.5       Equipment     \$22.1     \$104.0       Subtotel     \$446.4     \$166.0       Subtotel     \$446.4     \$168.0       General Administration     \$28.7     \$13.0       Project Total     \$475.1     \$179.0       Dollar amounts are shown in thousands of dollars.     \$0.0       Other Resources     Dollar amounts are shown in thousands of dollars.       Comments:     Closeout budget for molecular genetics portion of Project 96191A.       One man month of program manager is budgeted in the field portion of the 96191A project.		<u>it</u> -							
Budget Category:       FFY 1995       FFY 1996         Personnel       \$176.9       \$38.3         Travel       \$22.3       \$4.2         Contractual       \$22.3       \$4.2         Contractual       \$22.3       \$4.2         Subtotal       \$22.1       \$0.0         Example       \$22.1       \$0.0         Subtotal       \$24.7       \$13.0         General Administration       \$28.7       \$13.0         Project Total       \$445.4       \$166.0         Edurated       Estimated       Estimated         Estimated       Subtotal       \$445.4         General Administration       \$2.87       \$13.0         Project Total       \$475.1       \$179.0       \$0.0       \$0.0         Comments:       Dollar amounts are shown in thousands of dollars.       \$0.0       \$0.0       \$0.0         Comments:       Closeout budget for molecular genetics portion of Project 96191A.       One man month of program manager is budgeted in the field portion of the 96191A project.       FORM 3A         AGENCY       Project Tule: Investigating and Monitoring Oil Related Egg and Alevin Mortalities       AGENCY         Agency: AK Dept. of Fish & Game       Detail       B/3/95		Authorized	Proposed	and a second	da n an 'n che an na channan an uit annanan		and the second	an a	
Personnel       \$176.9       \$38.3         Travel       \$22.3       \$4.2         Contractuel       \$21.2       \$19.5         Equipment       \$2.1       \$0.0       LONG RANGE FUNDING REQUIREMENTS         Subtotal       \$44.64       \$166.0       Estimated       Estimated       Estimated         General Administration       \$28.7       \$13.0       FFY 1997       FFY 1998       FFY 1999       FFY 2000       FFY 2001       FFY 2002         Project Total       \$4475.1       \$179.0       Dollar amounts are shown in thousands of dollars.       \$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       \$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       \$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       \$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.	Budget Category:	FFY 1.995	FFY 1996						
Personnel       \$ 170.9       \$38.3         Travel       \$22.3       \$4.2         Contractual       \$212.9       \$104.0         Commodities       \$22.2       \$19.5         Equipment       \$2.1       \$0.0       LONG RANGE FUNDING REQUIREMENTS         Subtotal       \$24.6.4       \$166.0       Estimated       Estimated       Estimated         General Administration       \$28.7       \$13.0       FY 1997       FY 1999       FY 2001       FY 2001         Project Total       \$475.1       \$179.0       FY 1997       FY 1999       \$0.0       \$0.0       \$0.0         Collear amounts are shown in thousands of dollars.       Dollar amounts are shown in thousands of dollars.       Other Resources	Descent		100.0						
Investigation       \$22:3       \$42.3       \$42.3       \$42.3       \$6104.0         Commodities       \$32.2       \$19.5       LONG RANGE FUNDING REQUIREMENTS         Subtotal       \$42.3       \$106.0       Estimated       Estimate	Personnel	\$176.9	\$38.3						
Definition       \$2/2.3       \$104.0       Subtrail         Equipment       \$2.2       \$105.0       LONG RANGE FUNDING REQUIREMENTS         Subtrail       \$446.4       \$166.0       Estimated       Estimated       Estimated         General Administration       \$22.7       \$13.0       FFY 1997       FFY 1998       FFY 1999       FFY 2001       FFY 2002         Project Total       \$475.1       \$179.0       FFY 1997       FFY 1998       FFY 2000       \$0.0       \$0.0       \$0.0         Gther Resources		\$22.3	\$4.2						
Dominatiles       3.2.2       \$19.5       ION GRANGE FUNDING REQUIREMENTS         Subtorial       \$446.4       \$166.0       Estimated       Estimated </td <th></th> <td>\$212.9</td> <td>\$104.0</td> <td></td> <td></td> <td></td> <td></td> <td></td> <td></td>		\$212.9	\$104.0						
Equipment       \$2.1       \$0.0       LONG RANGE FONDING REQUIREMENTS         Subtotal       \$446.4       \$166.0       Estimated		\$32.2	\$19.5		ali akanta alima a manta mata ing tang maning kana ang maning sa				
Subtotal ⁴ /446.4 <u>\$106.0</u> ⁴ /46.4 <u>\$106.0</u> ⁵ /28.7 <u>\$113.0</u> FFY 1998 FFY 1998 FFY 1999 FFY 1990 FFY 2002 FFY 2002 FFY 2002         FFY 2002 FFY 2002 FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FFY 2002         FORM 34         FORM 34         AGENCY         Project Title: Investigating and Monitoring Oil Related Egg and Alevin         Mortalities         Agency: AK Dept. of Fish & Game         FFY 2002         FFY 202         FORM 34         AGENCY         PROJECT         DETAIL         Bi/3/95         Bi/3/95         FORM 34         AG	Equipment	\$2.1	\$0.0		LONG	RANGE FUNDIN	IG REQUIREMEN	NTS	
General Administration       \$28.7       \$13.0       FFY 1997       FFY 1998       FFY 1999       FFY 2000       FFY 2001       FFY 2002         Project Total       \$475.1       \$179.0       \$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       \$0.0       \$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       \$0.0       \$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       \$0.0       \$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       \$0.0       \$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       \$0.0       \$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       \$0.0       \$0.0 <td< td=""><th>Subtotal</th><td>\$446.4</td><td>\$166.0</td><td>Estimated</td><td>Estimated</td><td>Estimated</td><td>Estimated</td><td>Estimated</td><td>Estimated</td></td<>	Subtotal	\$446.4	\$166.0	Estimated	Estimated	Estimated	Estimated	Estimated	Estimated
Project Total       \$475.1       \$179.0       \$0.0       \$0.0       \$0.0       \$0.0         Full-time Equivalents (FTE)       1.0       Dollar amounts are shown in thousands of dollars.       Other Resources       State	General Administration	\$28.7	\$13.0	FFY 1997	FFY 1998	FFY 1999	FFY 2000	FFY 2001	FFY 2002
Full-time Equivalents (FTE)       1.0         Dollar amounts are shown in thousands of dollars.         Orments:         Closeout budget for molecular genetics portion of Project 96191A.         One man month of program manager is budgeted in the field portion of the 96191A project.         Project Number: 96191A - Interim Molecular Color	Project Total	\$475.1	\$179.0				\$0.0	\$0.0	\$0.0
1.0       1.0       Dollar amounts are shown in thousands of dollars.         Other Resources       Dollar amounts are shown in thousands of dollars.         Comments:       Closeout budget for molecular genetics portion of Project 96191A.         One man month of program manager is budgeted in the field portion of the 96191A project.         Project Number: 96191A - Interim Malecular Genetics         Project Title: Investigating and Monitoring Oil Related Egg and Alevin Mortalities         Agency: AK Dept. of Fish & Game									
Dollar amounts are shown in thousands of dollars.         Comments:         Closeout budget for molecular genetics portion of Project 96191A.         One man month of program manager is budgeted in the field portion of the 96191A project.         Image: state of the state	Full-time Equivalents (FTE)		1.0	le com d'a tetristications an tim-an actuation dans	an anti-anticity and a state of the state of the state of the	and an and a state of the second of the seco	an air aine an an an the air an	it is approximation of any provident the	en e
Uther Hesources				Dollar amoun	ts are shown ir	thousands of o	dollars.		
Comments: Closeout budget for molecular genetics portion of Project 96191A. One man month of program manager is budgeted in the field portion of the 96191A project. One man month of program manager is budgeted in the field portion of the 96191A project. Project Number: 96191A - Inferior Molecular Genetics Project Title: Investigating and Monitoring Oil Related Egg and Alevin Mortalities Agency: AK Dept. of Fish & Game Project Murber: 96191A - Inferior Molecular Genetics Project Title: Investigating and Monitoring Oil Related Egg and Alevin Mortalities Agency: AK Dept. of Fish & Game	Other Resources				<u> </u>	1		· · · · · · · · · · · · · · · · · · ·	
<b>1996</b> Project Number: 96191A - Interim Molecular Genetics       FORM 3A         AGENCY       Project Title: Investigating and Monitoring Oil Related Egg and Alevin       AGENCY         Mortalities       PROJECT       DETAIL         Prepared:       1 of 5       8/8/95	One man month of program	manager is budgeted	l in the field po	ortion of the 96	191A project.				
Mortalities       PROJECT         Agency:       AK Dept. of Fish & Game       DETAIL         Prepared:       1 of 5       8/8/95	Project Number: 96191A - Interim Molecular Genetics Project Title: Investigating and Monitoring Oil Related Egg and Alevin AGE					FORM 3A AGENCY			
Prepared: 1 of 5 Agency: AK Dept. of Fish & Game DETAIL 8/8/95		Mortalities							PROJECT
Prepared: 1 of 5 8/8/95		Agency: AK	Dept. of Fish	& Game					DETAIL
	Prepared: 1 of	5						L	8/8/95

# 1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

Pers	onnel Costs:		GS/Range/	Months	Monthly		Proposed
PM	Name	Position Description	Step	Budgeted	Costs	Overtime	FFY 1996
*	D. Moore J. McMahon Vacant	Program Manager Fish and Wildlife Technician II Fish and Wildlife Technician II	18L 9C 9B	0.0 9.0 3.0	6,333 3,217 3,100	0 0 0	0.0 29.0 9.3
	I	<u>Cubtotal</u>		12.0	12 650		
Tho	se costs associated with prog	Sublotal	ment of an *	12.0	12,050	U Personnel Total	\$38.3
Tray	val Costs	an management should be indicated by place	Ticket	Round	Total	Doily	Proposed
PM	Description		Price	Trins	Davs	Per Diem	FFY 1996
						,	
	Attend scientific meeting		800	1	5	95	1.3
	Anchorage/Cordova - Juneau	(Attend meeting with NMFS per.)	444	2	6	95	1.5
	Project planning and impleme	mtation.	224	3	8	95	1.4
Tho	se costs associated with prog	ram management should be indicated by place	ement of an *.	•••••••••••••••••••••••••••••••••••••••		Travel Total	\$4.2
<b>1996</b> Project Number: 96191A         Project Title: Investigating and Monitoring Oil Related Egg and Alevin         Mortalities         Agency: AK Dept. of Fish & Game						FORM 3B Personnel & Travel DETAIL	
	2 of 5						8/8/95

2 of 5 L

# 1996 EXXON VALDEZ TRU: : COUNCIL PROJECT BUDGET

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October 1, 1995 - September 30, 1996

Contractual Costs:		Proposed
Description		FFY 1996
DNA equipment repair (\$3.6) and maintenance agreements (\$1.0)		4.6
Anchorage wet lab repair and maint. Air charter (Little Port Walter/Sitka) 2 RT @ \$ 0.5/RT Contract with New York University Medical Center for sequencing RNA transcriptors. Contract with University of Alaska Anchorage to support 9 mos. of post. doc. to evaluate genetic lesions using DNA sequencing.		2.0 1.0 50.5 45.9
When a non-trustee organization is used, the form 4A is required.	al Totali	\$104.0
Commodities Costs:		Proposed
Description		FFY 1996
Laboratory chemicals (\$6.5) and supplies (\$2.2)		8.7
Lab equipment repair and maintenance parts		2.0
Anchorage wet lab supplies (\$3.3), lab chemicals (\$4.0) and supplies (\$1.5)		8.8
Commoditie	s Total	\$19.5
<b>1996</b> Project Number: 96191A Project Title: Investigating and Monitoring Oil Related Egg and Alevin Mortalities 3 of 53 of 5Agenoy: AK Dept. of Fish & Game	F Co Co	ORM 3B ntractual & mmodities DETAL5

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

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

New	Equipment Purchases:		Number	Unit	Proposed
Des	cription		of Units	Price	FFY 1996
		ι;			
Tho	se purchases associated with	replacement equipment should be indicated by placement of an R.	New E	quipment Total	\$0.0
Exis	ting Equipment Usage:			Number	Inventory
Des					Agency
	1996	Project Number: 96191A Project Title: Investigating and Monitoring Oil Related Egg and Ale Mortalities Agency: AK Dept. of Fish & Game	evin		FORM 3B Equipment DETAIL
	5 of 5	L	·····	J	8/8/95
### 1996 EXXON VALDEZ TRUSILE COUNCIL PROJECT BUDGET October 1, 1995 - September 30, 1996

		Authorized	Proposed						
Budget Category:		FFY 1995	FFY 1996						
Personnel			\$120.4						
Travel			\$120.4						
Contractual			\$45.3						
Commodities			\$12.4						
Equipment			\$2.1		LONG F		G REQUIREMEN	NTS	
Subtotal			\$189.3	Estimated	Estimated	Estimated	Estimated	Estimated	Estimated
General Administration			\$21.2	FFY 1997	FFY 1998	FFY 1999	FFY 2000	FFY 2001	FFY 2002
Project Total			\$210.5	\$167.9	\$167.9	\$46.2	\$0.0	\$0.0	\$0.0
		F							
Full-time Equivalents (F	TE)		2.2						
				Dollar amount	s are shown in	thousands of c	ollars.	· · · ·	
Other Resources									
Costs for FY96 and be monitoring will continu out only.	yond are e: e to show	stimated assumi no difference in	ng (1) that the embryo mortali	incubation expe	eriment will be o	discontinued af d sites for three	ter the 1995 br additional year	rood year and ( rs. FY99 costs	2) that field are for close

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

D	and al Calibra			L			<b>1</b> • •
Pers			GS/Kange/	Months	Monthly	<b>•</b> •	Proposed
PM		Position Description	Step	Budgeted	Costs	Overtime	FFY 1996
ľ	Vacant	Program Manager	18L	1.0	6,333	C	6.3
	M. Willette	Fishery Biologist III	18D	2.0	5,915	0	· 11.8
	A. Craig	Fishery Biologist I	14A	8.0	4,144	1,300	34.5
	Vacant	6 - Fish and Wildlife Technician II & III	11A	10.0	3,992	C	39.9
	D. Evans	Biometrician II	19C	4.0	6,095	C	24.4
	P. Trautman	Field Office Assistant	11A	1.0	3,509	C	3.5
							0.0
							0.0
							0.0
							0.0
	L						0.0
		Subto	tal	26.0	29,988	1,300	
Tho	se costs associated with pro	ogram management should be indicated by pla	cement of an *.		P	ersonnel Tota	I <u>\$120.4</u>
Tra	vel Costs:		Ticket	Round	Total	Daily	Proposed
PM	Description		Price	Trips	Days	Per Dien	FFY 1996
ļ.	Attend biometrics consulta	tion and planning mtg.	200	9	13	95	5 3.0
	Attend pink and chum wor	kshop	500	3	6	95	5 2.1
	Attend meeting with NMFS	S personnel	500	3	9	95	5 2.4
	Attend state AFS meeting		400	3	4	95	5 1.6
I							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
							0.0
The	se costs associated with pro	ogram management should be indicated by pla	cement of an *.			Travel Tota	<u>\$9.1</u>
	<u></u>						
		Project Number: 961914					FORM 3B
1					A las dim		Pareonnal
	1996	Project little: Investigating and Mo	nitoring Uli Rela	ited Egg and	Alevin		reisonnel
		Mortalities					& Iravel
		Agency: AK Dept. of Fish & Game	9				DETAIL

### 1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

Contractual Costs:			Proposed
Description			FFY 1996
Air charter for sample transport (1	2 hours @ \$0.25/hour)		3.0
Vessel charter for Fall Embryo San	npling (R/V Montague @ \$1.2K/day for 25 days)		' 30.0
Air charter for Fall Embryo samplir	ng (12 hours @ \$0.25/hour)		3.1
D.O.T. vehicle rental (2 months @	\$0.3/month)		1.2
Air charter for transport of person	nel/samples from field monitoring and hatchery rearing experiments (16 hours @ \$0.2	5/hr.	4.0
Hatchery space rental and room a	nd board for incubation sampler (\$1.0/month x 3.5 months)		3.5
Air charter (Little Port Walter/Sitka	1) 2 RT @ \$ 0.5/RT		1.0
Cordova outboard (\$0.5), UV bact	ericidal depurators (\$1.0), egg incubator (\$1.0), repair and maintenance agreement		2.5
		×	
When a non-trustee organization is	s used, the form 4A is required.	Contractual Total	\$45.3
Commodities Costs:			Proposed
Description			FFY 1996
Laboratory chemicals (\$2.0) and s	upplies (\$0.7)		2.7
Data processing supples			1.5
Lab equipment repair and mainten	ance parts		1.5
Egg dig related field sampling sup	olies (\$2.8), AFK hatchery wetlab supplies (\$3.9)		6.7
	A for a second se		
· · ·	ίτατα τη διατική τη από τη		
			[
		<b>Commodities Total</b>	\$12.4
······································			
	Project Number: 96191A		FORM 3B
	Destant Titles Investigation and Maritaging Oil Delated Francis d Al.		ntractual &
1996	Project litie: investigating and Monitoring Uli Related Egg and Alevin		
	Mortalities		ommodities
	Agency: AK Dept. of Fish & Game		DETAIL
3 of 4			7/31/95

#### 1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

130004 TO October 1, 1995 - September 30, 1996

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New	/ Equipment Purchases:						Number	Unit	Propose
Des	cription	Month	Esstion				of Units	Price	FFY 199
Des	Fry pump for field mon Replacement outboard	VitimoM itering)(Com motor (25 h) 007,1 4 1,6 7 20,2 9.002 1 20,2 1 20,2	(antroot pontent A) p) for field (0 S I (0 B (0 C (0 C (0 C (0 C) (0 C) (0 C) (0 C) (0 C) (0 C)	- <u> </u>		and the second s	of Units	Price	FFY 199 0.5 0.6 0.6 0.6 0.6 0.6 0.6 0.6 0.6 0.6 0.6
	<b>41</b> 1		1.0		12.1		oshnydDj		915 1.CT 0.0 0.0
Tho	se purchases associated	with replace	ement equipme	ent should be	indicate	d by placement of an R.	New E	quipment Tota	\$2.1
Exis	ting Equipment Usage: :	P.		្រាល់ ហ្	16-445	weet should be wanted by new	STABLE NYSTOP	they oNumber	an Inventor
Des	cription	15 231	lteus			an	Mark Lada, Lada Naja da	of Units	Agenc
	Hydraulic fry pumps		12915 '	un terreter anti-anti-anti-anti-anti-anti-anti-anti-			strapstan in	4	ADF
	a -2- * ≩ -21 €	le-			•	.se⊂ PMPS bec}	ः २.२१ <b>९९१९ ५</b> ) २.२१४ - २१४ - २२४४	nan sina 1 San Aragotti si 1 San sec	1000 1000 1000
		· · {		•••					
	1996	Proj Proj Mor Age	ect Number ect Title: In rtalities ency: AK De	: 96191A ivestigating ept. of Fish	and N & Gar	Monitoring Oil Related Egg an me	d Alevin		FORM 3B Equipment DETAIL

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1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 1995 - September 30, 1996

						1775 N. 1976 No. 18	12
Pers	onnel Costs:		GS/Range/	Months	Monthly	· · · · · · · · · · · · · · · · · · ·	Proposed
PM	Name	Position Description	Step	Budgeted	Costs	<b>Overtime</b>	1996 FFY 1996
	M. Willette	Fishery Biologist III	18D	2.0	5,915	t rangang 9.40	11.8
	Vacant	Fishery Biologist II	16A	12.0	4,700	0	56.4
	A. Craig	Fishery Biologist I	1.4A	8.0	4,144	1,300	34.5
	Vacant	Data Analyst	18A	1.0	5,354	0	5.4
	Vacant	6 - Fish and Wildlife Technician II & III	11A	12.5	3,992	0	49.9
	D. Evans	Biometrician I	17C	5.0	6,095	0	30.5
	P. Trautman	Field Office Assistant	11A	1.0	3,509	0	3.5
*	PCN 117064	Program Manager	18L	0.5	6,333	0	3.2
	J. McMahon	Fish and Wildlife Technician II	9C	12.0	3,217	0	38.6
	Vacant	Fish and Wildlife Technician II	9B	3.0	3,100	0	9.3
*	PCN 116110	Librarian I	17J	0.3	5,530	0	1.7
	1	Subto	tal <b>di si si si si si</b>	57.3	51,889	1,300	
Tho	se costs associated wi	ith program management should be indicated by pla	cement of an *.		F	Personnel Total	\$244.8
Trav	vel Costs:		Ticket	Round	Total	Daily	Proposed
PM	Description		Price	Trips	Days	Per Diem	FFY 1996
	Anchorage-Cordova (	(Planning mtg./ biometrics consulation)	224	6	13	95	2.6
	Attend scientific mee	nting	500	3	6	95	2.1
	Anchorage/Cordova -	- Juneau (Attend meeting with NMFS per.)	444	3	9	95	2.2
	Project planning and	implementation.	224	6	12	95	2.5
	Anchorage - Cordova	(AFK hatchery experiments)	224	15	30	95	6.2
Tho	se costs associated wi	ith program management should be indicated by pla	cement of an *.			Travel Total	\$15.6
		Project Number: 961914	ı				FORM 3B
l		Droject Titley Investigation and May		ad Passand .			Porconnol
1	1996	Project fille: investigating and Mol	nitoring Oil Relate	ed Egg and /	Alevin		
1		Mortalities					& Iravel
		Agency: AK Dept. of Fish & Game	)				DETAIL
	2	2 of 4					5/2/95

#### 1996 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

#### October 1, 1995 - September 30, 1996

Contractual Costs:					Proposed
Description				······································	FFY 1996
Air charter for sample 1	transport (S	12 hours @ \$0.25/hour)			23.0
DNA equipment repair	(\$3.6) and	maintenance agreements (\$1.0)			4.6
Vessel charter for Fall I	Embryo Sa	npling (R/V Montague @ \$1.1K/day for 23 days)			25.3
Air charter for Fall Emb	oryo sampli	ng (12 hours @ \$0.25/hour)			3.0
D.O.T. vehicle rental (2	2 months @	\$0.3/month)			0.6
Air charter for transpor	rt of person	nel/samples from field monitoring and hatchery rearing experiemnts (16	hours @ \$0.25/hr.		4.0
Hatchery space rental a	and room a	nd board for incubation sampler (\$1.0/month x 3.5 months)			3.5
Anchorage wet lab repa	air and mai	nt.			2.0
Air charter (Little Port \	Walter/Sitk	a) 2 RT @ \$ 0.5/RT			1.0
Contract with New Yor	rk Universit	y Medical Center for sequencing RNA transcriptors.			50.5
Hatchery space rental a	and room a	nd board for sampler			1.2
Contract with Washing	ton State L	Iniversity for deleterious mutation work			15.0
Cordova outboard (\$0.	5), UV bac	tericidal depurators (\$1.0), egg incubator (\$1.0) repair and maintenance a	agreement	، ۱	2.5
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When a non-trustee org	ganization i	s used, the form 4A is required.	Co	ntractual Total	\$136.2
Commodifies Costs:	15460 A 1	BINE BEARD, BREEDLIGUE RECEARE L. AND PORCE DA LARCEMENTE OF			Proposed
Description		ու որ հանձանությունը հայտությունը հանձանական հանձանությունը է հայտությունը է հայտությունը է է է է է է է է է է է Այս հայտությունը հայտությունը հայտությանը հանձանական է համարին է է է է է է է է է է է է է է է է է է է		No. of the second s	FFY 1996
Laboratory chemicals (	\$6.5) and s	upplies (\$2.2)			8.7
Data processing supplie	es (\$1.3) a	nd software (\$0.7)			2.0
Lab equipment repair a	nd mainten	ance parts		i	2.0
Field supplies, nets, rai	in gear, glo	ves, boots, hatchery supplies	1		3.0
Egg dig related field sa	mpling sup	olies (\$2.8), AFK hatchery wetlab supplies (\$3.9)			6.7
Anchorage wet lab sup	plies (\$3.3	), lab chemicals (\$4.0) and supplies (\$1.5)			8.8
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The second secon			Com	modities Total	\$31.2
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		Project Number: 96191A			TORM 3B
1006		Project Title: Investigating and Monitoring Oil Related Egg a	and Alevin	Co	ntractual &
1330		Mortalities	•	Co	mmodities
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	HOCK DATA AND AND AND AND AND AND AND AND AND AN		- Charles
New Equipment Purchases:	Numbe	r Unit	Proposed
Description		s Price	FFY 1996
Fry pump for field monit	oring (Component A)	n whites Intal	0.5
, and the set was a star	CARAGE STATE STATE STATE (CONTRACT)		
Econig related field same no	(二世)地方・マクロッド Note (1991C) 1901 - 20 January (2019)		4.4 A
Feur supplies, nots, rain gear	, gioves, boots, heldling calpulate		
Lating population repair cost ma	internorm parts		
Date woonsaing supplies (s)	[0] and software (50.7)		2
. Prodock obenhade (30.0)	and supplies (92 %)		
		rent and the second test	A0.5
Those purchases associated v	with replacement equipment should be indicated by placement of an R.	equipment rotal	\$0.5
Existing Equipment Usage:	To the form 1/ is regulad.		inventory
Description		of Units	Agency
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	Project Number: 96191A		ORM 3B
1006	Project Title: Investigating and Monitoring Oil Related Egg and Alevin		quinment
1996	Mortalities		
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L]	Agency. AN Dept. of Fish & Game		
4 of	4		5/2/95