

EXXON VALDEZ TRUSTEE COUNCIL
FY 94 DETAILED PROJECT DESCRIPTION

Project title: Stock Identification of Chum and Sockeye Salmon in Prince William Sound

Project ID number: 94137

Project type: Restoration/Monitoring

Name of project leaders: Samuel Sharr, Alaska Dept. Fish & Game
Carol J. Peckham, Alaska Dept. Fish & Game

Lead agency: Alaska Department of Fish and Game

Other Cooperating Parties: Prince William Sound Aquaculture Corp.
Valdez Fisheries Development Assoc.

Cost of project/FY 94: \$261.6
Cost of project/FY 95: \$261.6
Cost of Project/FY 96 and beyond:

Project Start-up/Completion Dates: 15 April 1993 - 15 March 1994

Geographic area of project: Prince William Sound

Project leader:



Sam Sharr (ADF&G) *By Brian Bue*

3/4/94

Date

Agency project manager:

Joe Sullivan (ADF&G)

Date

B. INTRODUCTION

Recent wild stock production in Prince William Sound (PWS) has ranged from 800 to 900 thousand chum salmon *Oncorhynchus keta* and 300 to 500 thousand sockeye salmon *O. nerka*. Up to 75% of wild pink and chum salmon spawn in intertidal areas. Oil from the *Exxon Valdez Oil Spill* (EVOS) which was deposited in these intertidal spawning areas adversely affected the survival of pink salmon embryos in 1989 and 1990 and subsequent generations from these populations have experienced reproductive impairment (Sharr et al. 1993a and 1993b). Pink salmon which exited PWS through oiled marine waters in 1989 also exhibited lower growth and survival than those which did not (Willette and Carpenter 1993). Although virtually all salmon damage assessment research has been conducted on the numerically superior pink salmon, it is likely that other salmon species were similarly damaged. Chum salmon share intertidal spawning areas with pink salmon and both sockeye and chum salmon share migratory corridors with pink salmon and were therefore exposed to oil in a similar manner. The suite of injuries already identified for pink salmon may have led to recent declines in the overall well being of wild pink salmon populations. It is believed that these effects may persist for several years and may apply to other species as well. Returning sockeye and chum salmon are older than pink salmon and post spill estimates of their survival are not complete. The full extent of damage to these species is not yet known.

Salmon populations impacted by the EVOS are heavily exploited in commercial, sport, and subsistence fisheries. These populations can most effectively be restored through stock-specific management practices designed to reduce exploitation on impacted wild populations. Wild salmon returns from areas heavily impacted by the EVOS are present in greatest numbers in large mixed stock commercial fisheries dominated by huge hatchery returns. Because their early life stages occur in a protected environment hatchery populations are potentially more productive and less variable than wild populations and did not sustain injury from the EVOS during the embryo stage. Wild salmon populations originate from hundreds of streams and lakes around PWS. Their migratory timing and abundance in marine fishing areas varies, and so to sustain their productivity managers must insure that adequate numbers of fish from all portions of the return escape fisheries and enter streams to spawn. Exploitation rates in mixed stock fisheries must be limited to levels appropriate for the least productive stock present at the time. To implement restoration premised on such stock-specific management, we must be able to distinguish wild fish from hatchery fish, and identify temporal and spatial abundance trends for each in commercial fisheries.

This project is designed to provide accurate and precise real-time catch contribution estimates for hatchery and wild salmon stocks in Prince William Sound. Accurate escapement estimates from ongoing Alaska Department of Fish and Game (ADF&G) escapement monitoring projects will enable managers to identify stocks which are experiencing escapement shortfalls. Accurate and timely catch contribution estimates from this coded wire tag recovery project will enable managers to identify times and areas where exploitation on depleted wild populations can be minimized and still permit the harvest of surplus hatchery returns. Post season analyses of the catch contribution estimates together with results from salmon escapement enumeration projects will provide stock-specific estimates of total return and survival and enable managers to assess the effectiveness of stock-specific management strategies.

In the absence of improved stock-specific management capabilities afforded by this project, salmon populations in western PWS which have already been stressed and depleted by the oil impacts may be over-exploited in the commercial, sport and subsistence fisheries. Injured wild spawning populations may be reduced to levels below that required for rapid recovery and in some instances, virtual elimination of a stock. If adequate population monitoring programs are not in place, changes in fishing effort to areas of less oil impact could also result in over exploitation of otherwise healthy, uninjured wild stocks. The feasibility of using coded wire tags to estimate catch contributions of hatchery produced salmon in PWS was thoroughly investigated by ADF&G from 1984 through 1988. During the EVOS damage assessment process, large scale tagging and recovery projects were instituted and perfected by ADF&G Natural Resources Damage Assessment (NRDA) Fish/Shellfish (F/S) Study #3. Damage assessment funds were expended to tag hatchery releases of sockeye, chum, coho, and chinook salmon in 1989 and 1990. NRDA funds were also used to tag sockeye salmon in the outmigrations from Coghill and Eshamy Lakes in 1989, 1990, and 1991 and from Jackpot Lake in 1990 and 1991. Tag recovery efforts for wild and hatchery salmon were funded by damage assessment funds in 1989, 1990, and 1991 and by Restoration funds in 1992 and 1993. Some age classes of stocks which were tagged with EVOS Trustee Council funds will return in 1994. If recovery efforts proposed here are not instituted in 1994 important restoration and population monitoring data will be irretrievably lost.

Results of this study will provide estimates of hatchery and wild stock contributions to commercial harvests, hatchery cost recovery harvests and hatchery brood stocks. Stock-specific catch contributions by fishing district and date will be used inseason by fisheries managers to reduce effort on injured wild stocks and target effort on healthy hatchery returns. Post season analyses of current year, as well as historic tag recovery data, will be coupled with escapement data for wild stocks to make estimates of wild stock total returns. Post season analyses of tagging data will also identify trends in the temporal and spatial distributions of stocks in the fisheries. These data are important for fisheries managers who must anticipate the effects of fishing strategies in future years if depleted stocks are to be protected. Protection of less productive stocks through stock-specific fisheries management has been successful in previous years. Stock-specific management strategies which incorporated data from coded-wire tagging projects funded by the Natural Resources Damage Assessment and Restoration processes were used successfully in the past to justify time and area fishery closures and effectively reduce exploitation on oiled stocks in mixed stock fisheries in 1990, 1991, 1992, and 1993. Serious escapement shortfalls were avoided despite intense fishing pressure on surplus hatchery fish in adjacent areas.

C. PROJECT DESCRIPTION

1. Resources and/or Associated Services:

This restoration and monitoring project is designed to provide estimates of hatchery and wild sockeye and chum salmon contributions to commercial and cost recovery fisheries in PWS, AK. These estimates will allow fisheries managers to lessen interceptions of wild fish in mixed stock fisheries. The project will be administered and supervised by ADF&G.

2. Relation to Other Damage Assessment/Restoration Work:

The foundations for this project were firmly established during the pre-EVOS era by coded wire tagging and recovery studies conducted initially by ADF&G and later cooperatively by ADF&G and two private non-profit aquaculture associations in PWS, Prince William Sound Aquaculture Corporation (PWSAC) and Valdez Fisheries Development Association (VFDA). The study initially began in 1984 with the tagging of chum salmon releases from the ADF&G Main Bay Hatchery (MB) (Attachment I). ADF&G, PWSAC, and VFDA cooperative studies began in 1986 and continued through 1988. These studies included tagging of hatchery releases of pink salmon from Armin F. Koernig (AFK), Wally Noerneberg (WN), Cannery Creek (CC), and Solomon Gulch (SG) hatcheries (Attachment I). Chum salmon from SG, sockeye salmon from MB, and coho salmon from SG and WN were also tagged. Following the March 24, 1989 EVOS the cooperative study was continued as part of the EVOS NRDA Fish/Shellfish (F/S) Study #3, a large scale code wire tagging and recovery project for hatchery and wild salmon in PWS. In 1989 this project tagged representative portions of all pink, sockeye, and coho salmon release from PWS hatcheries, chum salmon releases from SG, and wild sockeye salmon migrating out of Coghill and Eshamy Lakes. In 1990 and 1991, tagging efforts were expanded to include hatchery chum salmon releases from WN, hatchery releases of chinook salmon, and wild sockeye salmon migrating out of Jackpot Lake. Although EVOS funding for tag application was discontinued by the Trustees in 1991, area Aquaculture Associations have continued to apply tags to all their releases. EVOS funding for tag recovery has continued to the present.

The EVOS Trustee Council has also funded a Coghill Lake Sockeye Salmon Restoration Project (94259). That study is designed to investigate and restore the salmon rearing habitat in Coghill Lake but cannot succeed if adequate spawning escapement is not achieved through stock specific fisheries management.

3. Objectives:

- a. Make inseason estimates of the temporal and spatial contributions of tagged hatchery stocks of sockeye and chum salmon to PWS commercial and hatchery harvests based on the number of tags detected in adipose clipped fish which are recovered during catch sampling;
- b. provide timely inseason estimates of hatchery and wild stock contributions to harvests by time and area to fisheries managers so they can closely regulate exploitation of injured wild stocks;
- c. use data from fully decoded tags recovered from commercial catches, cost recovery harvests, and hatchery brood stock to verify or adjust inseason contribution estimates and;
- d. estimate marine survival rates for each uniquely coded hatchery release and wild outmigration group where possible.

4. 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 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 Catches

The Alaska Department of Fish and Game will oversee the recovery of coded-wire tagged fish in commercial salmon harvests in Prince William Sound. The recovery samples will be from a stratified sample. Recoveries will be stratified by district and fishing period. Recoveries will also be stratified by processor based on the finding of Peltz and Geiger (1988) who detected significant differences between proportions of some tag codes among sampled processors. These differences indicate that processors receive catches from only certain portions of a district or fishing period stratum. It is believed that this effect is the result of tendering and fish buying practices specific to each processor.

For each time and area specific stratum, 25% of the sockeye and chum salmon catch will be scanned for fish with a missing adipose fin. Each fish sampled will be subject to visual and tactile examination for a missing adipose fin denoting the presence of a coded wire tag. Recoveries of tags in chum and sockeye salmon are made on conveyor belts as fish are unloaded from tenders or on fish gutting and heading lines in four processing facilities located in Cordova, one facility in Whittier, three facilities in Valdez and one facility in Anchorage. When feasible, samplers for the pink salmon coded wire tag recovery project (94-184) stationed at facilities in Kodiak, Kenai and Seward and on large floating processors will also sample sockeye and chum salmon harvests from PWS. All deliveries by fish tenders to these facilities will be monitored by radio and by daily contact with processing plant dispatchers to ensure that the catch deliveries being sampled are district-specific.

Data recorded for each tender sampled will include harvest type, (ie., commercial or cost recovery catch), fishing district where the catch was made, catch date, processor, number of fish examined, and number of clips observed. The total number of fish aboard each tender and for each sampled spatial and temporal stratum are obtained later from an ADF&G computerized record of processor sales receipts (fish tickets). Heads of adipose-fin clipped fish will be excised, identified with a uniquely numbered cinch tag, bagged, and frozen. Samplers at locations outside of Cordova or Valdez will air-freight data and heads semi-weekly to the Cordova office. Project biologists will carefully inventory, edit, and log all sampling data as it is returned to the Cordova and Valdez offices.

Hatchery Sales Harvests

In addition to catch sampling at the processing facilities, approximately 40% of the fish in the hatchery terminal harvest areas will be scanned for fish missing adipose fins. Because sales harvests are processed at the same processing facilities as commercial harvests, methods and means of sampling sales harvest will be identical to those described for commercial catches.

Hatchery Brood Stocks

Brood stock sampling is critical to estimating hatchery and wild contributions. Due to differential mortality between tagged and untagged fish as well as differential tag loss between release groups, the tag expansion factor at release for hatchery fish may not accurately reflect the tag expansion factor in the adult population. Hatchery brood stocks will be scanned for tags in order to estimate adjustment factors which could 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. Although there is evidence that the first assumption is not valid for pink salmon (Sharr et al. 1993a) there is no similar evidence to invalidate it for chum or sockeye salmon. 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 hatchery. 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 for chum salmon in Sharr et al. (1993 a), and some more direct preliminary evidence is discussed by Sharr et al. (1993c) with respect to pink salmon but in neither case is the evidence conclusive to completely invalidate the third assumption. At present all three assumptions are assumed to be true for chum and sockeye salmon.

The adjustment factor 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. For fish which mature at different ages such as chum and sockeye salmon the adjustment factor must be specific to a release year. The adjustment factor for hatchery h and release year y will be estimated as the ratio of sampled fish from release year y in the brood stock to the expanded number of fish from release year y based on tags found in the sample:

$$\hat{a}_{hy} = \frac{\hat{s}_{hy}}{\sum_i \frac{x_i}{p_i}}, \quad (1)$$

where

- T_y = number of tag codes released from hatchery h , in release year y ,
 p_i = tagging rate at release for i th tag code released in year y (defined as number of tagged fish released with i th tag code divided by the total number of fish in release group i),
 s_{hy} = number of brood stock fish examined in hatchery h from release year y and,
 x_i = number of tags of i th tag code released in year y , found in s_{hy} .

The untagged portion of s_{hy} will be estimated using age data derived from representative scale and otolith samples taken from the broodstock. The adjustment factor will then be used to adjust contribution estimates (Equation 3) if it can be shown that it was significantly greater than 1.0 at the 90% level. An appropriate test of the hypothesis :- $H_0 : a_{hy} \leq 1.0$, given in Sharr *et al* (1993 b), will be modified to account for the variability contributed by the estimation of s_{hy} .

There will be a brood stock tag recovery effort at each of the three hatchery facilities where tags were initially applied to sockeye and chum salmon. Technicians will be stationed at each of these hatcheries to scan the brood stock during egg take. After the salmon are manually spawned, technicians will use visual and tactile methods to scan approximately 95% of the fish. Total number of fish scanned and total number of fin-clipped fish found will be recorded on a daily basis. Representative random samples of paired otolith and scale samples will be taken daily to estimate the numbers of fish from each release year y in the brood stock. Scale and otolith sample sizes will be large enough to estimate proportion of each release year within ± 5 percentage points of the true proportion 95% of the time. Heads excised from fish with missing adipose fins, sampling data sheets, and scale and otolith samples, will be picked up on a regular basis and returned to Cordova for editing and processing.

Wild Escapements

One hundred percent of the wild escapement through ADF&G operated management weirs at Coghill and Eshamy lakes will be sampled for tags. Representative portions of the brood will be sampled for scales, sex and size as part of the ADF&G management program (Wilcock *et al.* 1993). Adjustment factors for these escapements will be calculated as described for hatchery broodstocks.

Tag Extraction, Tag Decoding, and Data Archiving

During the fishing season all sampling data and frozen heads from fish adipose-clipped fish will be sent daily to the ADF&G Coded Wire Tag Processing Laboratory in Juneau (Tag Lab). Data received at the Tag Lab will be logged and tag recovery sampling forms edited a second time for accuracy and completeness. Samples which affect critical fisheries decisions are prioritized and heads from those samples will be processed immediately. Tag Lab staff will locate and remove tags from heads, decode extracted tags, and enter tag code and sample data into a statewide coded wire tag database. 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 Prince William Sound 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:

$$\hat{C}_t = \sum_{i=1}^L x_{it} \left(\frac{N_i a_{hy}}{s_i p_t} \right), \quad (2)$$

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_{hy} = adjustment factor for hatchery h and release group y .

L = number of recovery strata associated with common property, cost-recovery, brood stock, special harvests and escapement in which tag code t was found.

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

$$\hat{C}u_t = \sum_{j=1}^U N_j * \left(\frac{\sum_{j=1}^S \hat{C}_{tj}}{\sum_{j=1}^S N_j} \right), \quad (3)$$

where

U =number of unsampled strata,

N_j =number of fish in j th unsampled stratum

S =number of strata sampled in the period in which the unsampled stratum resides,

C_{tj} =contribution of release coded with tag t to the sampled stratum j , and

N_j =number of fish in j th sampled stratum.

When a district-week opening 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 Prince William Sound return for 1994 will be obtained through summation of contribution estimates for sampled and unsampled strata. An estimate of the total hatchery contribution to the Prince William Sound 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:

$$\hat{V}(\hat{C}_t) = \sum_{i=1}^L x_{it} \left[\frac{N_i \hat{a}_{hy}}{s_i p_t} \right] \left[\frac{N_i \hat{a}_{hy}}{s_i p_t} - 1 \right]. \quad (4)$$

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 strata are believed to be small (Sharr et al., 1993).

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

$$\hat{S}_t = \frac{\hat{C}_t + \hat{C}u_t}{R_t}, \quad (5)$$

where

C_t = contribution of release coded with tag t to sampled strata,

Cu_t = 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 \hat{S}_t is given by:

$$\hat{V}(\hat{S}_t) = \frac{\sum_{h=1}^L x_h \left[\frac{N_t \hat{a}_h}{s_t p_t} \right] \left[\frac{N_t \hat{a}_h}{s_t p_t} - 1 \right]}{R_t^2}. \quad (6)$$

In-Season Hatchery Contributions

Inseason fisheries decisions which must be made on very short notice require rapid, real time analysis of coded wire tag data. Inseason contribution will be estimates will be made according to the same procedures outlined for post-season estimates except that the mean of historical adjustment factors for each hatchery will be used because current year brood stock data are not available until the end of the season.

Contributions of Tagged Wild Populations

Contribution and survival estimates for tagged wild salmon will be derived in a manner similar to those for tagged hatchery fish (equations 6 and 8) as will be the variances of the contribution and survival rates (Equations 7 and 9). An estimate of the contribution of the release group coded with wild stock tag code t to the total PWS return will be the summation of contribution estimates from the commercial and cost recovery harvests and from the wild escapement.

Alternatives

Estimation of stock specific contributions to large commercial fisheries requires some sort of natural or man-induced mark which 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 for accurate and precise estimates of catch contributions, (4) not affect survival or behavior of fish and, (5) application and or recovery of the mark must be cost effective.

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 cost effectiveness of coded wire tag technology decreases as the size of populations marked increases and the size of the fish marked decreases. The pink salmon tagging program in PWS is the largest of it's kind in the world and is pushing the limit of the technology for both application and recovery. Furthermore, application in very small fish such as pink and chum salmon may effect survival, may not be permanent (tag loss), and tagging may affect behavior. Some cost effective methods do exist and are used to adjust for tag loss from differential mortality and tag shedding. However, the effect of tag loss from tag-induced straying, though thought to be small, is difficult and costly to account for.

Based on the drawbacks of coded wire tag technology with respect to PWS wild salmon, an alternative mark or method for distinguishing stocks from one another would certainly be desirable. The most likely alternative to coded wire tags are thermal otolith marks. This marking method meets all of the five criteria described above. Thermal marks have been thoroughly tested in all salmon species. The marks are permanent, they are easily applied to every individual in a hatchery population and they 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 and 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 1994 (Study 94187). Recoveries of otolith marks from these releases can begin in 1996.

To date no natural markers have been discovered in PWS chum salmon which allow researchers to distinguish hatchery stocks from all wild stocks. Scale patterns have proven useful for one wild stock of sockeye salmon have nor been used successfully to distinguish other wild sockeye salmon stocks from hatchery returns. 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 hence, are probably not distinguishable. Creating a genetic mark in hatchery populations by selective breeding is a possibility which is under consideration but would take several generations to develop given the size of hatchery production in PWS.

5. Location:

By aiding restoration through improved fisheries management this project will benefit wild sockeye and chum salmon populations in PWS and other segments of the marine and terrestrial portions of the PWS ecosystem which are dependent upon them. Restoration through improved management will also benefit the salmon fishing fleets including those in

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, and Anchorage 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.

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 Coast 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 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.

7. Contracts:

None.

D. SCHEDULES

Date(s)	Activity
June 20 - Sept 10, 1994	Tag recoveries in commercial fisheries, cost recovery harvests, and brood stocks. Inseason catch stock composition estimates by time and area for management of commercial and cost recovery fisheries.
Nov 15, 1994	Draft Report
Dec 30, 1994	Final Report

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), hire project personnel, supervise day to day project operations, maintain data quality, assist in data analyses, and coauthor final reports. The APL will be assisted by two non-permanent Fisheries Biologist I (FB I). One 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 PWSAC hatcheries. Another FB I will supervise day to day sampling activities in Valdez and at the VFDA hatchery. A non-permanent Fish and Wildlife Technician III (FWT III) will assist the Cordova FB I and act as a crew leader. Fish and Wildlife Technician II's (FWT II) will be placed in ports where only one sampler is expected to work under minimum supervision. The remainder of crews in each port will be pairs of non-permanent FWT II's and Fish and Wildlife Tech I's (FWT I). Each day, one or two persons will scan salmon at each processing plant. Under the supervision of the FB I, a FWT III in Cordova will conduct daily data logging, editing and archiving activities. The second FB I will supervise similar activities in 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. 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.

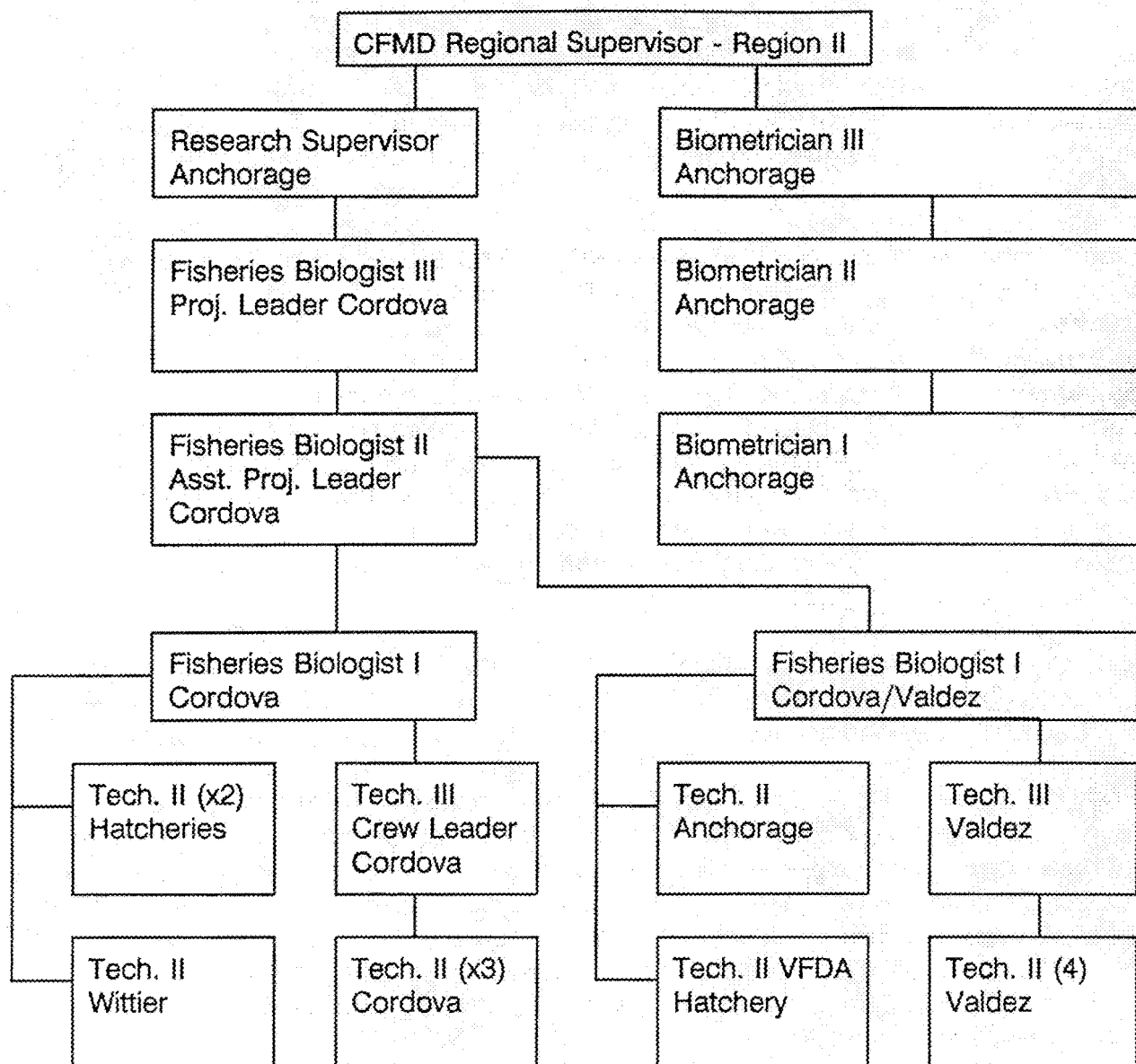


Figure 1. Organizational chart of project personnel and related ADF&G CFMD Region II supervisory and biometrics staff.

E. EXISTING AGENCY PROGRAM

The Alaska Department of Fish and Game permanent staff of biologists and biometricians write operational plans and provide overall all supervision for this project. Prince William Sound Aquaculture Corporation and Valdez Fisheries Development Association apply tags to all species reared in their hatchery facilities. 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.

F. ENVIRONMENTAL COMPLIANCE/PERMIT/COORDINATION STATUS

All sampling activities for this project occur within fish processing plants of fish hatcheries. The ADF&G will coordinate with PWSAC and VFDA with respect to locating samplers within their respective fish hatcheries.

G. PERFORMANCE MONITORING

The PL is the permanent full time Fisheries Biologist III PWS Salmon Research project Leader for the ADF&G, Region II, Commercial Fisheries Management and Development Division (CFMD). The PL is supervised by the ADF&G, CFMD, Region II, Regional Research Supervisor. The APL is an eleven month permanent season ADF&G employee supervised by the PL. The APL has supervisory authority of all project Fisheries Biologist I's and 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 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, one of the Project Fisheries Biologist I's 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 I and the Biometrician II. Technician III crew leaders with the project can be replaced in the short term by the Fisheries Biologist I's. Several Technician II's have been with the project for more than one season and qualify as easily trained replacements.

The PL is a permanent ADF&G employee and can work on project tasks at any time. The APL is seasonal but dedicated 11 months to project tasks. Fisheries Biologist I's who assist the APL will assume their duties in early June approximately two weeks prior to the onset of recovery activities for pink salmon. Fish and Wildlife Technicians employed for sampling will be hired in mid-June and assume their duties when commercial or cost recovery fisheries commence. Crews sampling in Anchorage, Whittier, and Valdez will be hired locally and will provide their own room and board. Project biologists will visit each port and minimum of once every two weeks to answer questions, and provide quality control supervision.

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, 1993. 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. Incomplete censusing of tender loads of fish from fish offloading conveyors will be tested for randomness when the opportunity for an independent total census arises. All data are edited immediately upon completion of sampling and are edited twice more by Statewide tag laboratory personnel.

The project has real time in-season as well as longer term post-season products. Real time estimates of wild and hatchery catch contributions will be used to make inseason fisheries management decisions. Data for inseason estimates will be from important segments of the fishery which have the biggest impact on wild salmon populations. Samples from these strata will be given top priority. Data sheets will be edited and logged and together with heads and copies of the data log, shipped to the Juneau Tag Lab. Tag laboratory personnel cross check all samples received with the accompanying copy of the data log and work around the clock 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 provide fisheries managers with reliable catch stock contribution estimates.

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 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 Prince William Sound 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.

H. COORDINATION OF INTEGRATED RESEARCH EFFORT

This project is intimately integrated with the pink salmon coded wire tag recovery project (94184). The two projects are fully integrated with respect to a common project leader, assistant project leaders, biometrics support, sampling personnel, Tag Lab personnel, and all project logistics. Both coded wire tag projects will also integrate tender fleet tracking, processor plant logistics, and crew scheduling with an existing ADF&G salmon port sampling projects. Local aquaculture associations which apply provide all tagging, fry release, sales harvest, and brood stock data necessary for data analysis. Aquaculture associations also provide room,

board, and logistics support for brood stock 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.

The monitoring, research and restoration objectives of this project are very related to ecosystem based research and restoration efforts described in part by the Sound Ecosystem Assessment (SEA) plan. Although the SEA project and a related projects focus to some extent on pink salmon, pink salmon have many early life history attributes in common with chum and sockeye salmon. Chum salmon share the same intertidal stream rearing habitat as pink salmon and both chum and sockeye salmon share the migratory corridors used by chum salmon. It is likely sockeye and chum salmon share many of the same food resources as pink salmon and are preyed upon by the same predator populations. Therefore, some study results for pink salmon may be applicable to sockeye and chum salmon and it is also likely that considerable data about chum and sockeye salmon will be collected in as part of SEA studies of pink salmon and their ecology. Without the release group survival estimates from this study, interpreting and taking advantage of this early life history data for the other species of salmon will not be possible. Therefore, this project is related to several other projects including the suite of SEA projects (94320), the Salmon Otolith Marking (94187), and the Pink Salmon Egg and Alevin Mortality (94191) projects. These estimates are critical to several components of SEA including those investigating:

- 1) salmon survival in relation sea surface temperature and other oceanographic features of PWS during the fry and juvenile life stages.
- 2) salmon survival in relation to abundance, size, growth rate, and distribution of pink salmon fry and juveniles and, zooplankton population distribution, abundance, and species composition, and
- 3) salmon survival in relation to abundance, size, growth rate, and distribution of salmon fry and juveniles and the abundance distribution, size, and species composition of predator populations.

This project is also directly linked to the proposed Otolith Marking project (94187). Otolith marking is a logical extension of marking technology which will ultimately replace many of the functions of coded wire tags and provide more accurate and precise estimates of hatchery and wild contributions to salmon catches and escapements in PWS at less expense. However, until otolith marks can be applied, coded wire tagging and recovery projects will continue to provide those estimates.

I. PUBLIC PROCESS

The general public has been involved in the development and evolution of the coded wire tag program in Prince William Sound since its inception in the early 1980's 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 coded-wire tag projects have been 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.

J. PERSONNEL QUALIFICATIONS

Fisheries Biologist III Project Leader - Samuel Sharr

Mr. Sharr received a Bachelor of Science degree in biology from the University of Washington in 1968. He has been a research biologist for ADF&G since 1979 and has worked on PWS salmon and herring since 1981. He assumed his present position as the ADF&G, Division of Commercial Fisheries, Biologist III, PWS Area Fin Fish Research Project Leader in 1986. In this capacity, Mr. Sharr oversees all the salmon and herring research conducted by the Division of Commercial Fisheries in PWS. His involvement with the PWS salmon escapement aerial survey program dates from the early 1980's. Mr. Sharr has supervised a total re-edit of the historic aerial and ground survey data and designed a new RBASE data base for inseason escapement analyses. Mr. Sharr wrote the original operational plans for NRDA F/S Studies 1,2 and, 3, in 1989 and 1990, and 1991, restoration studies 60A, 60B, and 60C in 1992, and 93137, 93184, and 93191 in 1993 and has been the Principal Investigator for all of those projects. Mr. Sharr is also a member of the scientific committee of the Prince William Sound Fisheries Ecosystem Planning Group and a co-author of the Sound Ecosystem Assessment research plan and science proposal.

Fisheries Biologist II Assistant Project Leader - Carol Peckham

Ms. Peckham has a Bachelor of Science in Wildlife Biology from the University of Alaska and complete all course work requirements for a Masters degree in statistics. She has been employed by ADF&G since 1984. As a college intern for the ADF&G Stock Biology Group Ms. Peckham gained valuable experience in a wide variety of biological sampling and stock identification techniques in Cook Inlet and Prince William Sound. Ms. Peckham is been involved in coded-wire tag recovery activities in PWS since their inception and since 1987 she has been the Fisheries Biologist in charge of coded-wire tag recovery operations for PWS salmon. Her experience includes supervision of sampling activities spread throughout south central Alaska. She has co-authored several reports in the ADF&G Technical Data Report series, interim and final reports for the 1991 NRDA F/S Study #3, the 1992 Restoration Study 60C, and 1993 Restoration studies 93137 and 93184.

Biometrician I - David Evans

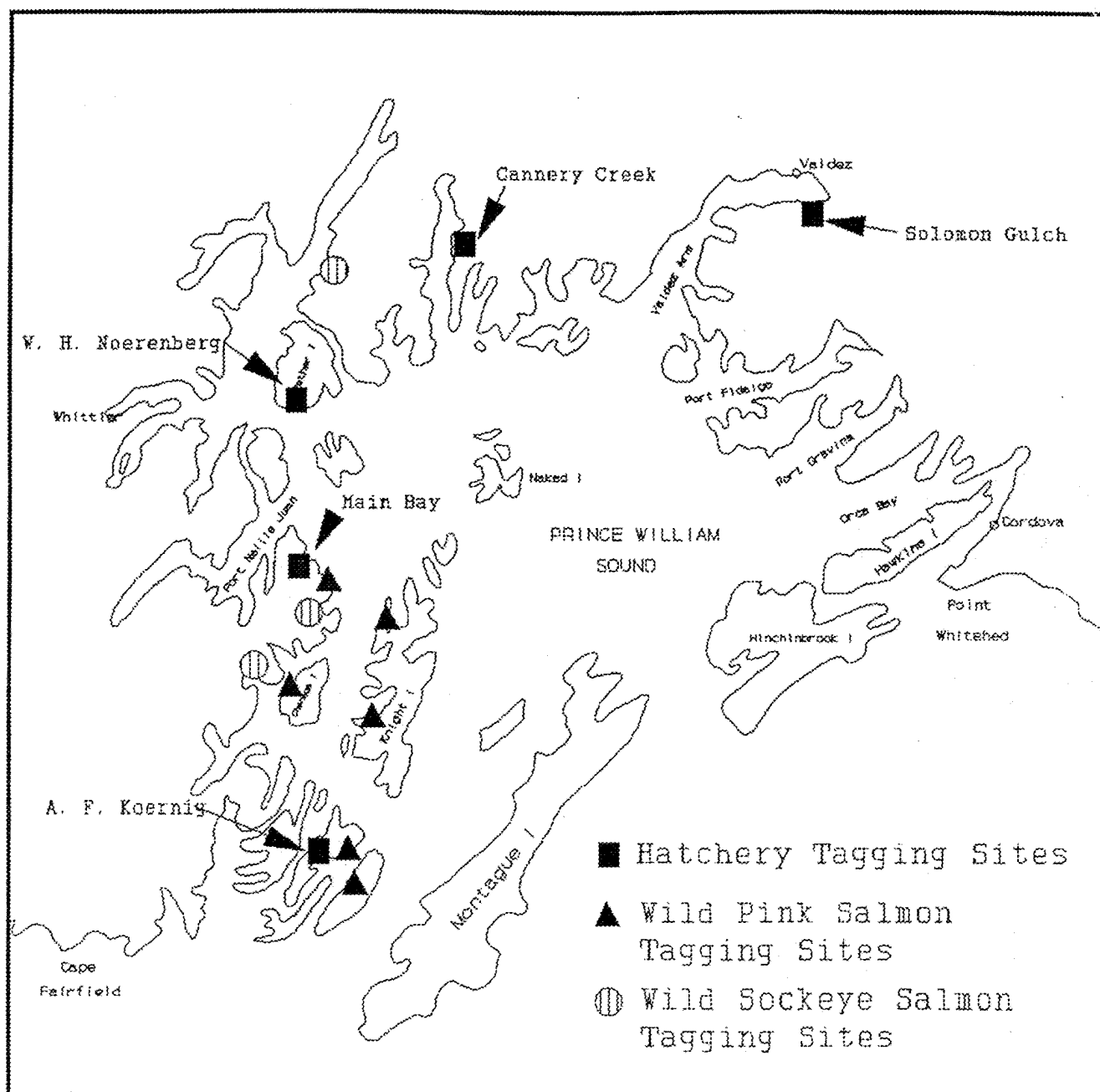
David Evans has a Bachelor of Science in soil science from the University of Nottingham (U.K.), a Master of Science and a Doctor of Philosophy degree in soil science from the University of Guelph (Ontario, Canada), and a Master of Science in statistics from Oregon

State University. David has worked with the Alaska Department of Fish and Game since October, 1991. His primary responsibility has been analysis of coded-wire-tag data from Prince William Sound. He has designed the statistical procedures and computer spread sheets used for inseason analysis of tag recovery data, has overseen most of the post season data analyses and has co-authored interim and final reports for the 1991 NRDA F/S Study #3, the 1992 Restoration Study 60C, and 1993 Restoration studies 93137 and 93184.

K . BUDGET (attached)

LITERATURE CITED

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- Wilcock, J.A.. 1993. Salmon Catch and Escapement Statistics For Copper River, Bering River, and Prince William Sound, 1989. Alaska Dept. of Fish and Game, Commercial Fisheries Management and Development Division, Technical Fishery Report 93-07, Juneau, Ak.



Attachment 1. Map showing the location of tagging sites for Hatchery and wild stocks of salmon which will contribute to adult returns in 1992.

EXXON VALDEZ TRUSTEE COUNCIL
1994 Federal Fiscal Year Project Budget
October 1, 1993 - September 30, 1994

Project Description: Stock Identification of Chum, Sockeye, Chinook & Coho in PWS - This project recovers coded wire tags from adult sockeye, chum, chinook and coho salmon tagged as fry at four hatcheries in Prince William Sound. It makes estimates of wild and hatchery catch contributions, total returns, and survival rates. Inseason catch contribution estimates for hatchery and wild fish permit fisheries managers to modify time and area fishing patterns to protect oil damaged wild pink salmon stocks.

Budget Category:	1993 Project No. 93068 Authorized FFY 93	'93 Report/ '94 Interim* FFY 94	Remaining Cost** FFY 94	Total FFY 94	FFY 95	Comment
Personnel	\$105.8	\$36.1	\$163.8	\$199.9	\$208.9	The FFY 95 budget may vary depending on results from FFY 94.
Travel	\$1.1	\$0.8	\$1.0	\$1.8	\$1.8	
Contractual	\$2.8	\$3.2	\$17.2	\$20.4	\$17.2	
Commodities	\$0.6	\$1.0	\$6.0	\$7.0	\$1.2	
Equipment	\$0.0	\$0.0	\$0.0	\$0.0	\$0.0	
Capital Outlay	\$0.0	\$0.0	\$0.0	\$0.0	\$0.0	
Subtotal	\$110.3	\$41.1	\$188.0	\$229.1	\$229.1	
General Administration	\$16.1	\$5.6	\$26.9	\$32.5	\$32.5	
Project Total	\$126.4	\$46.7	\$214.9	\$261.6	\$261.6	
Full-time Equivalents (FTE)	2.7	0.7	3.5	4.2	4.4	
Dollar amounts are shown in thousands of dollars.						
Budget Year Proposed Personnel: Position Description		Reprt/Intrm Months	Reprt/Intrm Cost	Remaining Months	Remaining Cost	
Reprt	9 Fish & Wildlife technician II			25.0	\$91.5	
	2 Fisheries Biologist II & III	2.0	\$10.4	2.0	\$12.0	
	1 Fisheries Biologist I	1.0	\$4.9	3.5	\$17.1	
	2 Analyst Programmers II & IV	0.4	\$1.9	0.8	\$3.8	
	1 Biometrician	1.0	\$5.0	0.0	\$0.0	
	1 Publication Specialist	0.2	\$0.9	0.4	\$1.8	
	2 Fisheries Technicians II/III (tag lab)	3.0	\$9.6	9.0	\$30.9	NEPA cost: \$0.0
	1 Program Manager	0.5	\$3.4	1.0	\$6.7	*Oct 1, 1993 - Jan 31, 1994
	Personnel Total	8.1	\$36.1	41.7	\$163.8	**Feb 1, 1994 - Sep 30, 1994

07/14/93

1994

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Project Number: 94137
Project Title: Stock ID of Chum, Sockeye, Chinook & Coho in PWS
Agency: AK Dept. of Fish & Game

FORM 2A
PROJECT
DETAILS

EXXON VALDEZ TRUSTEE COUNCIL
 1994 Federal Fiscal Year Project Budget
 October 1, 1993 - September 30, 1994

Travel:		Reprt/Intrm	Remaining
Reprt 2 RT between Anchorage & Cordova @ \$350/trip + \$50 per diem		\$0.8	\$0.0
Supervisory trips to Whittier (\$100), Kodiak (\$250), & Anchorage (\$350) + 2 days per diem @ \$150/day		\$0.0	\$1.0
Travel Total		\$0.8	\$1.0
Contractual:			
Intrm:	Air charter to hatcheries	\$2.0	
	DOT fleet vehicle	\$1.2	
Remain	Air charter to hatcheries, Valdez		\$7.0
	Office rental (Valdez, Whittier)		\$2.0
	DOT fleet vehicles		\$3.2
	Tag Lab head and supply shipments		\$4.0
	Tag Lab computer repair		\$1.0
Contractual Total		\$3.2	\$17.2

07/14/93

1994

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Project Number: 94137
 Project Title: Stock ID of Chum, Sockeye, Chinook & Coho in PWS
 Agency: AK Dept. of Fish & Game

**FORM 2B
 PROJECT
 DETAIL**

EXXON VALDEZ TRUSTEE COUNCIL
 1994 Federal Fiscal Year Project Budget
 October 1, 1993 - September 30, 1994

Commodities:		Reprt/Intrm	Remaining
Reprt	Professional and scientific supplies (rain gear, gloves, knives, etc.)	\$1.0	
	Rain gear, gloves, knives, sampling kits, supplies		\$2.0
	Tag lab supplies		\$4.0
Commodities Total		\$1.0	\$6.0
Equipment:			
Equipment Total		\$0.0	\$0.0

07/14/93

1994

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Printed: 3/10/94 12:02 PM

Project Number: 94137
 Project Title: Stock ID of Chum, Sockeye, Chinook & Coho in PWS
 Agency: AK Dept. of Fish & Game

FORM 2B
 PROJECT
 COUNCIL

94139A1

94139A1

**DETAILED PROJECT DESCRIPTION
UNDER PREPARATION FOR THIS PROJECT**

94139B1

**DETAILED PROJECT DESCRIPTION
UNDER PREPARATION FOR THIS PROJECT**

**DETAILED PROJECT DESCRIPTION
UNDER PREPARATION FOR THIS PROJECT**

94139C1

**DETAILED PROJECT DESCRIPTION
UNDER PREPARATION FOR THIS PROJECT**

94139C2

94139C2

**DETAILED PROJECT DESCRIPTION
UNDER PREPARATION FOR THIS PROJECT**

EXXON VALDEZ TRUSTEE COUNCIL
FY 94 DETAILED PROJECT DESCRIPTION

A. COVER PAGE

Project title: Survey to Monitor Marine Bird and Sea Otter Populations in Prince William Sound during Winter 1994

Project ID number: 94159

Project type: Restoration Monitoring

Name of principal investigator(s): Beverly A. Agler

Lead agency: U.S. Department of the Interior, Fish and Wildlife Service

Cooperating agencies: None


Cost of project/FY 94: \$188,600


Cost of project/FY 95: \$260,000

Cost of Project/FY 96 and beyond: \$43,000

Project Start-up/Completion Dates: January 1, 1994/April 30, 1995

Geographic area of project: Prince William Sound

Principal Investigator:  **Date:** 5/31/94
Beverly A. Agler, Wildlife Biologist
DOI-FWS (Region 7)

Project Manager:  **Date:** 5/31/94
David B. Irons, Wildlife Biologist
DOI-FWS (Region 7)

B. INTRODUCTION

The waters and shorelines of Prince William Sound (PWS) support abundant marine bird and sea otter populations throughout the year (Isleib and Kessel 1973, Hogan and Murk 1982, Irons et al. 1988a). Potential injuries to marine birds from exposure to the *T/V Exxon Valdez* oil spill included, but were not limited to, death, changes in behavior, and decreased productivity. Post-spill studies, identical to the one proposed here, suggested that the population abundance of sea otters (Burns) and several marine bird species (Klosiewski and Laing ms) declined as a result of the oil spill. Using surveys by small boats, this project will collect additional information on the winter distribution and abundance of marine birds and sea otters in PWS. These post-spill data will be compared to data collected previously to ascertain trends in marine bird and sea otter distribution and abundance in PWS. This project will benefit restoration of PWS by determining whether populations that declined due to the spill are recovering and identifying which species are still of concern.

C. PROJECT DESCRIPTION

1. Resources and/or Associated Services:

Marine birds and sea otters (*Enhydra lutris*) are the targeted resources. Nearly 30,000 bird (Piatt et al. 1990) and 900 sea otter (DeGange and Lensink 1990) carcasses were recovered following the spill. Of these totals, 3,400 bird (Piatt et al. 1990) and 490 sea otter (DeGange and Lensink 1990) carcasses were from PWS. Based on modeling studies using carcass search effort, and population data, an estimated 300,000 - 645,000 marine birds were killed in PWS and the northern Gulf of Alaska by the oil spill (Ecological Consulting, Inc. 1991). The majority of these birds were murrelets (*Uria* spp.; Piatt et al. 1990). The number of sea otters killed in PWS by the spill was estimated to be 2,800 otters (Garrott et al. 1993). These estimates are probably low because they only include direct mortality occurring in the first five months after the spill and do not include chronic effects or loss of reproductive output.

The U.S. Fish and Wildlife Service conducted boat surveys of marine bird and sea otter populations in PWS in 1972-73 (Dwyer et al. 1976), 1984-85 (Irons et al. 1988b), and several years following the spill (1989, 1990, 1991, and 1993; Klosiewski and Laing ms, Agler et al. ms). These surveys documented overall declines in PWS marine bird populations between the early 1970's (Dwyer et al. 1976) and the years after the spill. Species and species groups found to decline (Klosiewski and Laing ms) were cormorants (*Phalacrocorax* spp.), scoters (*Melanitta* spp.), black oystercatcher (*Haematopus bachmani*), scaup (*Aythya* spp.), glaucous-winged gull (*Larus glaucescens*), black-legged kittiwake (*Rissa*

tridactyla), Arctic tern (*Sterna paradisaea*), pigeon guillemot (*Cepphus columba*), *Brachyramphus* (marbled and Kittlitz's) murrelets (*B. marmoratus* and *brevirostris*), and northwestern crow (*Corvus caurinus*). Differences were also detected between the oiled area and non-oiled area. Nine species or groups (cormorants, harlequin duck (*Histrionicus histrionicus*), black oystercatcher, pigeon guillemot, northwestern crow, loons (*Gavia* spp.), mew gull (*Larus canus*), Arctic tern, and scoters) declined more in the oiled area (Klosiewski and Laing ms). Specific studies of three of these species - harlequin duck (Patton ms), black oystercatcher (Andres and Cody ms), and pigeon guillemot (Oakley and Kuletz ms) - corroborated the population changes found by the boat surveys.

Burn (ms), using data from the boat surveys, documented declines in sea otter abundance in shoreline habitats of PWS following the spill. He also detected a continuing pattern of significantly lower sea otter densities in oiled coastal areas, suggesting that mortality in or displacement of sea otters from these areas occurred.

Winter surveys were previously conducted in 1990, 1991, and 1993, but several more years of data collection are needed. Monte Carlo simulations using these data examined the estimated power of the abundance estimates. The simulations showed that power increased substantially for simulated sampling regimes in which data were collected every year rather than every other year over a 9-year period (Klosiewski and Laing ms).

This project has several benefits. Restoration of marine bird and sea otter populations requires population estimates to determine whether recovery is occurring or if declines are continuing. This project will benefit marine birds and sea otters by revealing species that show continuing injury due to the *T/V Exxon Valdez* oil spill. This information is necessary to plan meaningful restoration actions.

This project will also provide valuable information on the distribution and habitat use of these species. Survey data from this project have been used for these purposes by investigators of other studies on pigeon guillemots (G. Sanger, pers. comm.), marbled murrelets (K. Kuletz, pers. comm.), black oystercatchers (B. Andres, pers. comm.), and sea otters (Burn, ms). Survey methods are flexible enough to provide for collection of more detailed information (such as age class data) if such information is requested by other investigators.

2. Relation to Other Damage Assessment/Restoration Work:

Restoration of marine bird and sea otter populations requires population estimates to determine whether recovery is occurring or if declines are continuing. The Trustee council has supported three years of winter marine and sea otter populations (1990, 1991, and 1993). Survey data from this project have been used by investigators of other studies on marbled murrelets (K. Kuletz, pers. comm.), black oystercatcher (B. Andres, pers. comm), pigeon guillemots (G. Sanger, pers. comm.), and sea otters (D. Burn, pers. comm.).

3. Objectives

The purpose of this study was to obtain annual estimates of the winter population of marine birds and sea otters in PWS to monitor the recovery of species whose populations may have declined due to the *TV Exxon Valdez* oil spill. The specific objectives of this project are:

- a. To determine distribution and estimate population abundance, with 95% confidence limits, of marine bird and sea otter populations in Prince William Sound during March 1994;
- b. To determine whether the marine bird species whose populations declined more in oiled areas than in non-oiled areas of PWS have recovered;
- c. To examine the relative abundance of common species groups using data collected in March 1972, 1973, 1990, 1991, and 1993.
- d. To support restoration studies on harlequin duck, black oystercatcher, pigeon guillemot, marbled murrelet, and sea otters by providing data on population changes, distribution, and habitat use of PWS populations.

4. Methods

a. Study Area

Prince William Sound (approximately 60° 30' N, 147° W) is a large embayment of the northern Gulf of Alaska. The rugged coastline is dominated by the Chugach Mountains, which drop precipitously to the shoreline in an intricate pattern of fjords and bays. Including the mainland and more than 150 islands, PWS contains over 5000 km of shoreline. The depth of PWS varies from less than 1 fathom (2 m) on Middle Ground Shoal to more than 475 fathoms (870 m) east of Lone Island. The study area includes all water within PWS, as well as land within 100 m of the shoreline. The waters on the Gulf of Alaska side

of Montague, Hinchinbrook and Hawkins Islands, as well as Orca Inlet, are excluded.

b. Sampling Methods

Survey methodology will remain identical to that of post-spill surveys conducted in 1989, 1990, 1991, (Klosiewski and Laing ms) and 1993 (Agler et al. ms). For these surveys, we will use three 25-foot fiberglass boats, which are currently under U.S. Fish and Wildlife Service jurisdiction.

A stratified random sampling design containing 3 strata, shoreline, coastal-pelagic, and pelagic, will be used. The shoreline stratum includes all water within 200 m of shoreline, the strip of land within 100 m of the shore, and air space to an altitude of 100 m (Klosiewski and Laing ms). Irons et al. (1988a,b) divided the shoreline stratum of PWS into 742 transects. For the March survey, the number of transects are reduced to 13% of the total shoreline transects available because weather delays often extend the time required to complete the survey.

Both the coastal-pelagic and pelagic strata consist of plots of water delineated by 5-minute latitude and longitude intervals and exclude any water within 200 m of the coast. Coastal-pelagic plots intersect more than 1 nm (nautical mile) of shoreline, while pelagic plots intersect less than 1 nm of shoreline. Two north-south transect lines located 1 minute inside the east and west boundaries of each coastal-pelagic and pelagic plot are surveyed. When a plot is too small to contain two transects due to intersection with land, it is combined with an adjacent plot.

c. Data Analysis

As in previous surveys (Klosiewski and Laing ms, Agler et al. ms), we will use a ratio estimator (Cochran 1977) to estimate population abundance. Each stratum population for a given species or species group will be calculated as:

$$\hat{Y} = X\hat{R} \quad (1)$$

where:

\hat{Y} = population estimate for a stratum
 X = total area of all transects in the stratum

$$\hat{R} = \frac{\sum_{i=1}^n y_i}{\sum_{i=1}^n x_i}$$

Estimated variance of the population estimate will be calculated as follows:

$$\hat{V}(\hat{Y}) = X^2 \frac{N-n}{nN} \frac{1}{\bar{x}^2} \frac{\sum_{i=1}^n (y_i - \hat{R}x_i)^2}{n-1} \quad (2)$$

where:

$\hat{V}(\hat{Y})$ = estimated variance of \hat{Y}
 y_i = number of birds counted on i th transect
 x_i = area of i th transect sampled
 N = total number of transects in the stratum
 n = number of sampled transects in the stratum
 \bar{x} = mean area of all transects sampled in the stratum

For the coastal-pelagic and pelagic strata, there will be two deviations from the procedures shown. First, we will estimate y_i as the density of birds counted in transects multiplied by the area of the sampled block. Second, we will not include the finite population correction factor ($fpc = (N-n)/N$) shown in the variance formula (Formula 2) because the transect lines within plots are chosen systematically rather than randomly.

Population estimates for each species and species groups will be added to other post-oil spill population estimates to determine population trends. Regression analyses will be used to determine the recovery of injured species and population changes of other species.

d. Alternatives

We considered using aerial surveys as an alternative to boat surveys. Visibility bias is the main disadvantage to conducting marine bird surveys from the air. For example, in July 1989, boat-based observers surveyed 1/3 of the

PWS shoreline and counted 982 harlequin ducks; whereas, aerial observers counted only 705 for the total shoreline of PWS (Klosiewski and Hotchkiss ms). Accompanying this bias is a high variance in population estimates. Moreover, aerial surveys have typically required the same amount of time to complete because of flying limitations imposed by PWS weather, thus aerial surveys offer no temporal advantage.

5. Location

This study will be conducted in PWS. The study area includes all water within PWS, as well as land within 100 m of the shoreline including oiled and non-oiled areas. The waters on the Gulf of Alaska side of Montague, Hinchinbrook and Hawkins Islands, as well as Orca Inlet, are excluded.

6. Technical Support

All technical aspects, such as GIS, will be conducted by project personnel, therefore outside technical support is not needed.

7. Contracts

This project includes one contract of approximately \$18,000 for logistical support. A vessel large enough to provide lodging and meals for 9 people and 300 gallons of fuel per day will be chartered for logistical support and safety during the winter survey.

8. Literature Cited

- Agler, B. A., P. E. Seiser, S. J. Kendall, and D. B. Irons. ms. Marine bird and sea otter populations of Prince William Sound, Alaska: population trends following the *T/V Exxon Valdez* Oil Spill. Restoration Project No. 93045. Unpubl. Rep., U. S. Fish and Wildl. Serv., Anchorage, Alas. 60 pp.
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Klosiewski, S. P., and K. K. Laing. ms. Marine bird populations of Prince William Sound, Alaska, before and after the *Exxon Valdez* oil spill. NRDA Bird Study Number 2. Unpubl. Rep., U.S. Fish and Wildl. Serv., Anchorage, Alas. 85 pp.

Oakley, K. L., and K. J. Kuletz. ms. Population, reproduction and foraging ecology of pigeon guillemots at Naked Island, Prince William Sound, Alaska, before and after the *Exxon Valdez* oil spill. Bird Study Number 9. U.S. Fish and Wildl. Serv., Anchorage, Alas. 64 pp.

Patton, S. ms. Injury assessment of hydrocarbon uptake by sea ducks in Prince William Sound and the Kodiak archipelago, Alaska. NRDA Bird Study Number 11. Draft Report. Alaska Dept. Fish and Game, Anchorage, Alas.

Piatt, J. F., C. J. Lensink, W. Butler, M. Kendziorek, and D. R. Nysewander. 1990. Immediate impact of the 'Exxon Valdez' oil spill on marine birds. *Auk* 107:387-397.

D. SCHEDULES

1. FY1994 Milestones

Jan - Feb 94: Prepare Detailed Study Plan, hire personnel, make logistical arrangements for winter survey

Mar 94: Conduct winter survey in PWS

Apr 94: Return equipment and personnel to Anchorage, enter data, and clean and store equipment

Sept 94: Analyze data from March 1994 survey

2. FY1995 Milestones

Oct 94: Compile report of 1994 survey

Nov 30, 94: Draft Report to Oil Spill Coordinator

Jan 15, 95: Draft Report to Peer Review Committee

Apr 30, 95: Final Report complete

3. Project Personnel

JOB TITLE	GS LEVEL	MONTHS		RESPONSIBILITIES
		FY 94	FY 95	
Project Leader	GS-9/11	Jan - Sep	Oct - Feb	1) coordinating the collection of field data 2) timely reporting of data in draft and final reports
Biologist	GS-9	Mar - Aug	Oct - Feb	1) assist project leader
	GS-9	Mar - Aug		2) coordinates logistics for field surveys 3) organizes GIS database
Biological Technician	GS-5	Mar-Apr		1) participates in data collection
	GS-5			2) assists with logistics and preparation of equipment
	GS-5			3) performs data entry
	GS-5			
	GS-5			

4. Logistics

Surveys will require the use of three 25-foot vessels that are currently under U.S. Fish and Wildlife Service jurisdiction. In addition, a larger vessel will be chartered for fuel, lodging, and meals in remote areas during the winter survey.

E. EXISTING AGENCY PROGRAM

The Division of Migratory Bird Management of USFWS monitors reproduction of black-legged kittiwakes in PWS with base money. Without funding from the Trustee Council, winter seabird populations would not be monitored by USFWS. With approval of the forage fish studies in PWS, Project 94320 will be collecting data on forage fish (\$600 K). Project 94102 will be collecting data on murrelet foraging requirements (\$231.5 K), and Project 94173 will be the Pigeon guillemot recovery monitoring project (\$201.1K) in PWS.

F. ENVIRONMENTAL COMPLIANCE/PERMIT COORDINATION STATUS

This study relies on observations from boats and is a non-intrusive study. Based on a review of the CEQ regulation 40 CFR 1500-1508, this study has been determined to be categorically exempt from the requirements of NEPA in accordance with 40 CFR 1508.4.

G. PERFORMANCE MONITORING

1. Chain-of-command

Chief of Migratory Bird Management - Robert Leedy
Project Leader, Marine and Coastal Bird Project - Kenton D. Wohl
Project Manager, Marine Bird and Sea Otter Surveys- David B. Irons
Principal Investigator, Marine Bird and Sea Otter Surveys - Beverly A. Agler
Assistant Principal Investigator, Marine Bird and Sea Otter Surveys- Pam Seiser
Assistant Principal Investigator, Marine Bird and Sea Otter Surveys- Steve Kendall

The Assistant Principal Investigators and Biological Technicians report to the Principal Investigator. The Principal Investigator reports to Project Manager. The Project Manager is supervised by the Project Leader of the Marine and Coastal Bird Project, and the Project Leader is supervised by the Chief of Migratory Bird Management.

2. Backup Strategies

If personnel changes occur, replacements will be hired and trained accordingly. Despite personnel changes and other logistical obstacles, Migratory Bird Management has successfully conducted this type of survey for several years; therefore, have proven project time frames can be met.

3. Quality-assurance and Control Plan

To ensure that project design and procedures are followed, (1) all crew members will partake in training surveys prior to initial surveys, (2) one person on each boat will be responsible for maintaining consistent data collection procedures, (3) standardized forms will be used during data collection, (4) data forms will be checked at the end of each day to ensure the integrity of the data, and (5) all data entered into the computer will be carefully checked for errors. In addition, an observer manual and a transect guide were developed to maintain consistency in data collection over time and among observers. Use of global positioning systems (GPS) for finding and checking transect coordinates will help insure data accuracy as well.

H. COORDINATION OF INTEGRATED RESEARCH EFFORT

This project will provide valuable information on the distribution and habitat use of marine and coastal birds and sea otters in PWS. This project is being coordinated with other DOI-FWS and NBS seabird monitoring studies in PWS and elsewhere (ie., Lower Cook Inlet, Southeast Alaska). Survey data from this project will be available for use by investigators of other studies on harlequin ducks, marbled murrelets, black oystercatchers, and sea otters.

I. PUBLIC PROCESS

A work plan describing the proposed FY94 study was submitted to the Exxon Valdez Trustee Council in the fall of 1993, and this document was available to the public for review. The final report of this project will be available upon request to the general public. The private business sector are being given the opportunity to bid on the 1994 vessel contract work.

J. PERSONNEL QUALIFICATIONS

1. Principal Investigator - Beverly A. Agler, Wildlife Biologist, GS-9/11.

Beverly Agler received her M.S. degree in Wildlife Management from University of Maine, Orono in 1992 and her B.A. degree in Human Ecology from College of the Atlantic in 1981. Ms. Agler has worked for the U. S. Fish and Wildlife Service since May, 1993 as Project Leader of the Prince William Sound, Lower Cook Inlet, and Southeast Alaska population surveys of marine birds and sea otters. Prior to her arrival in Alaska, she participated in a joint National Science Foundation, National Oceanographic and Aeronautics Administration, University of Washington, and College of the Atlantic study of Antarctic seabirds and marine mammals. For over 10 years, she was the Project Director of the North Atlantic Fin Whale Catalogue, based at College of the Atlantic in Bar Harbor, Maine. She coordinated a collaborative study of fin whales in the western North Atlantic, including coordinating photographic identification of individuals, and genetic differentiation of individuals using skin biopsies. In addition to her work at College of the Atlantic, Ms. Agler was the Head Naturalist for Maine Whalewatch in Northeast Harbor, Maine. In this position, she led day-long offshore trips to study marine mammals and seabirds.

Selected Publications:

Agler, B. A. 1992. Photographic identification of individual fin whales (*Balaenoptera physalus*) in the Gulf of Maine. Master's Thesis, University of Maine, Orono. 157 pp.

- Agler, B. A. 1992. Testing the reliability of photographic identification of individual fin whales (*Balaenoptera physalus*). Rep. int. Whal. Commn. 42:731-7.
- Agler, B. A., and S. K. Katona. 1987. Photo-identification of finback whales. Whalewatcher 21:3-6.
- Agler, B. A., S. K. Katona, and B. Bowman. 1988. Maine's biggest wildlife mystery: a Maine endangered and nongame wildlife fund project report. Maine Fish and Wildlife 30:10-1.
- Agler, B. A., J. A. Beard, R. S. Bowman, H. D. Corbett, S. E. Frohock, M. P. Hawvermale, S. K. Katona, S. S. Sadove, and I. E. Seipt. 1990. Fin whale, (*Balaenoptera physalus*) photographic identification: methodology and preliminary results from the western North Atlantic. Rep. int. Whal. Commn (special issue 12):349-56.
- Agler, B. A., K. A. Robertson, D. DenDanto, S. K. Katona, J. M. Allen, S. E. Frohock, I. E. Seipt, and R. S. Bowman. 1992. The use of photographic identification for studying individual fin whales (*Balaenoptera physalus*) in the Gulf of Maine. Rep. int. Whal. Commn 42:711-22.
- Agler, B. A., R. L. Schooley, S. E. Frohock, S. K. Katona, and I. E. Seipt. 1992. Reproduction of photographically identified fin whales, *Balaenoptera physalus*, in the Gulf of Maine. J. Mamm. 74(3):577-87.
- Agler, B. A. In review. The effects of photographic matching errors population estimates of fin whales using capture-recapture data. Submitted to Rep. int. Whal. Commn.
- Agler, B. A., K. A. Robertson, and D. DenDanto. In review. The value of scars for the photo-identification of individual fin whales. Submitted to Can. J. Zool.

2. Project Manager - David B. Irons, Ph.D, Wildlife Biologist, GS-12.

David Irons received his Ph.D from the U. of CA, Irvine in 1992. His dissertation was on the foraging ecology and breeding biology of the black-legged kittiwake. The field work for this study was conducted in Prince William Sound. Irons received his M. S. from Oregon State University in 1982 where he studied foraging behavior of glaucous-winged gulls in relation to the presence of sea otters. Irons conducted marine birds and sea otter surveys in PWS in 1984 and 1985. He has been studying kittiwakes in PWS for 11 years and completed EVOS kittiwake damage assessment study. Irons has overseen several seabird studies in the past few years including a marine bird and sea otter survey in PWS and in Cook Inlet, a seabird monitoring study on Little Diomed Island, and a cost of reproduction study on kittiwakes. Irons has authored and co-authored several reports and publications on seabirds and has made several presentations at scientific conferences.

Selected Seabird Publications:

- Irons, D. B. 1992. Factors affecting black-legged kittiwake reproductive success. Unpubl. Ph.D Dissertation. Univ. of California, Irvine.

_____, R. G. Anthony, and J. A. Estes. 1986. Foraging strategies of glaucous-winged gulls in a rocky intertidal community. *Ecology* 67:1460-74.

_____. In review. Size and productivity of black-legged kittiwake colonies in Prince William Sound, Alaska before and after the *T/V Exxon Valdez* oil spill. Submitted to *Auk*.

_____. In prep. Foraging site fidelity and tidal rhythms in individual black-legged kittiwakes.

_____. In prep. Flexible foraging behavior in seabirds: short-term buffer and long-term tradeoff?

_____. In prep. The role of food availability in sibling aggression and brood reduction of the black-legged kittiwake.

Hatch, S. A., G. V. Byrd, D. B. Irons, and G. L. Hunt. 1993. Status and ecology of kittiwakes in the North Pacific Ocean. Pages 140-53 in K. Vermeer, K. T. Briggs, K. H. Morgan, and D. Siegel-Causey, eds. *The status, ecology and conservation of marine birds of the North Pacific*, Can. Wildl. Serv., Spec. Publ., Ottawa, Canada.

Hogan, M. E., and D. B. Irons. 1986. Waterbirds and marine mammals. Pages in M. J. Hameedi, and D. G. Shaw, eds. *Environmental management of Port Valdez, Alaska: scientific basis and practical results*. Springer-Verlag, New York.

Vermeer, K., and D. B. Irons. 1991. The glaucous-winged gull on the Pacific Coast of North America. *Acta Twentieth Congressus Internationalis Ornithologici*:2378-83.

K. BUDGET

Cost breakdowns for the FY94 restoration project are shown on Forms 2A and 2B. The cost shown for FY94 (\$188.6K) is the amount necessary to write the FY93 report (\$41.6K), conduct the winter survey (\$104.6), and prepare the FY94 report (\$42.4K).

EXXON VALDEZ OIL SPILL RECOVERY COUNCIL
1994 Federal Fiscal Year Project Budget
October 1, 1993 - September 30, 1994

Project Description: Surveys to monitor marine bird and sea otter populations on Prince William Sound. The project is designed to estimate the distribution and abundance of these species in both oiled and un-oiled areas of Prince William Sound.

Budget Category:	1993 Project No. 93075 Authorized FFY 93	'93 Report/ '94 Interim* FFY 94	Remaining Cost** FFY 94	Total FFY 94	FFY 95	Comment
Personnel	108.5	74.9	\$23.9	\$104.8		1993 Report
Travel	12.0	7.5	0.0	7.5		Personnel \$33.5
Contractual	80.0	23.0	0.0	23.0		Travel \$1.5
Commodities	10.0	9.0	0.5	8.5		Contractual \$1.0
Equipment	30.0	20.0	2.0	27.0		Commodities \$1.0
Capital Outlay	0.0	0.0	0.0	0.0		Equipment \$1.0
Subtotal	240.5	126.4	25.4	174.1		Capital Outlay \$0.0
General Administration	21.9	12.9	5.0	17.8		Subtotal \$30.5
Project Total	262.4	146.2	42.4	128.6		Gen. Admin. \$5.0
Full-time Equivalents (FTE)	3.4	2.4	0.5	1.4		Project Total \$41.0
Dollar amounts are shown in thousands of dollars.						
Budget Year Proposed Personnel:		Reprt/Intrm Months	Reprt/Intrm Cost	Remaining Months	Remaining Cost	FTE's
Position Description						
Reprt Principal Investigator Biologist		2.0	\$10.6	3.0	\$15.9	
Intrm Supervisory Biologist		0.5	\$3.0	0.5	\$3.0	
Biologist GS-9		2.0	\$11.0	2.0	\$11.0	
Biologist GS-9		2.0	\$8.0			
Biotechnician		3.0	\$6.8			
Expediter		1.0	\$2.0			
Personnel Total		10.5	\$41.4	5.5	\$29.9	
NEPA Cost: \$0.0						
*Oct 1, 1993 - Jan 31, 1994						
**Feb 1, 1994 - Sep 30, 1994						

07/14/93

1994

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Project Number: 94159
 Project Title: Marine Bird & Sea Otter Band Surveys
 Agency: Dept of Interior, Fish & Wildlife Service

**FORM 2A
 PROJECT
 DETAIL**

EXXON VALDEZ TRUSTEE COUNCIL
1994 Federal Fiscal Year Project Budget
October 1, 1993 - September 30, 1994

Travel:		Reprt/Intrm	Remaining
Reprt		7.5	
Intrm	Includes roundtrip to and from Prince William Sound via train, and cash for March 1994 Survey		
<u>March 94</u> Train - 6 trips x \$1000/trip = \$6,000 Truck - \$20/day x 30 days = \$600 Per diem - \$30/day/person x 10 people x 30 days = \$900			
Travel Total		\$7.5	0.0
Contractual:			
Reprt		\$18.0	
Intrm	Charter Vessel for March 1994 survey Training		
<u>Charter vessel</u> \$1,800/day x 10 days = \$18,000			
<u>Training</u> First aid/CPR - 10 people x \$100 each = \$1,000 Marine Survival - 10 people x \$150 each = \$1,500 Watercraft Safety - 10 people x \$150 each = \$1,500 Firearm/Bear Awareness - 10 people x \$100 each = \$1,000		\$5.0	
Contractual Total		\$23.0	0.0

07/14/93

1994

Page 1 of 2

Project Number: 94159

Project Title: Marine Bird & Sea Otter Boat Surveys

Agency: Dept of Interior - Fish & Wildlife Surveys

FORM 2B
PROJECT
D

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EXXON VALDEZ TEE COUNCIL
1994 Federal Fisca. Project Budget
October 1, 1993 - September 30, 1994

Commodities:			
Reprt			
Intm	Food for March 1994 survey, printer paper, miscellaneous expendable supplies for reports & repairs	\$9.0	
	<u>March 1994</u>		\$0.5
	Food - 10 people x \$25/day x 30 days = \$7,500		
	Expendables \$200		
	Printer materials \$150		
	Repairs on office equipment \$200		
	<u>Remaining</u>		
	Expendables for report \$200		
	Printer Cartridge \$100		
	Repairs on office equipment \$200		
	Commodities Total	\$9.0	\$0.5
Equipment:			
Reprt			
Intm	Winter Boat Storage, replacement gear, and boat maintenance and repairs	\$20.0	\$7.0
	<u>Intm</u> Winter storage / maintenance \$5,000		
	motor replacement 2x \$5,000 each \$10,000		
	Repairs/parts to prepare for survey \$2,000		
	Repairs/parts as needed during survey \$3,000		
	<u>Remaining</u>		
	Winterize boats 3x \$1,000 \$3,000		
	Replace/repair parts \$3,000		
	Clean boats \$1,000		
	Equipment Total	\$20.0	\$7.0

07/14/93

1994

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Project Number: 94151
Project Title: Marine Bird & Sea Otter Boat Surveys
Agency: Dept. of Interior; Fish & Wildlife Service

FORM 2B
PROJECT
DETAIL

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94163

EXXON VALDEZ TRUSTEE COUNCIL
FY 94 DETAILED PROJECT DESCRIPTION

Project title: Forage Fish Influence on Recovery of Injured Species

Project ID number: 94163

Project type: Research/Monitoring

Name of project leader(s): Bruce Wright, NOAA
David Irons, USFWS
Mark Willette, ADF&G

Lead agency: NOAA

Cooperating agencies: U.S Fish and Wildlife Service, USFWS
Alaska Department of Fish and Game, ADF&G

Cost of project/FY 94:

Cost of project/FY 95:

Cost of Project/FY 96 and beyond:

Project Start-up/Completion Dates: March 1, 1994 - September 30, 1994

Geographic area of project: Prince William Sound

Name of project leader:

Name of lead agency project manager:

B. INTRODUCTION

C. PROJECT DESCRIPTION

This is a multi-year project designed to estimate the degree of diet overlap among forage fish species in PWS. The carrying capacity of PWS for forage fish is a function of primary and secondary productivity and the degree of prey resource partitioning (Cooney 1993). Our present lack of knowledge regarding prey resource partitioning among forage fish limits our ability to estimate the carrying capacity of PWS (Cooney 1993). Prey resource partitioning among forage fish species is a function of the degree of habitat and diet overlap among species. The NOAA contractor will evaluate the degree of habitat overlap among forage fish species.

A pilot study will be conducted during the first year of the project to obtain data needed to refine sample size estimates, evaluate several analytical techniques for describing diet overlap and prey electivity, and determine if prey categories can be pooled in future years. In addition, we will test for differences in prey composition (abundance & biomass) and stomach fullness (% body weight) between two laboratories. The results from the first year of study will be used to refine the study design used in the future years.

1. Resources and/or Associated Services:

This project will focus on adult and juvenile fish that compose the prey resources (forage fish) used by apex predators (marine birds and mammals). Important forage fish species may include Pacific herring (*Clupea harengus pallasii*), Pacific sandlance (*Ammodytes hexapterus*), capelin (*Mallotus villosus*), walleye pollock (*Theragra chalcogramma*), Pacific cod (*Gadus macrocephalus*), and juvenile salmon (*Oncorhynchus spp.*). The information obtained from the study will also contribute to our understanding of the mechanisms affecting population dynamics of forage fish.

2. Relation to Other Damage Assessment/Restoration Work:

This project will complement the Sound Ecosystem Assessment (SEA) program. SEA is a multi-disciplinary program designed to develop an understanding of the mechanisms regulating ecosystem function in PWS. During its first year, SEA will focus on the interactions of pink salmon and herring with other components of the PWS ecosystem. The Salmon Predator and Salmon Growth and Mortality components of the SEA program will collect samples for this project during the May and June sampling periods (Table 1).

3. Objectives:

This project will achieve the following objectives during the 1994 season:

1. Collect samples of forage fish for analysis of stomach contents as well as available prey (zooplankton and epibenthic invertebrates) in three habitat types during four time periods.
2. Conduct laboratory analyses of epibenthic invertebrate/zooplankton samples and fish stomach contents.
3. Test for differences in the measurement of prey composition (abundance & biomass) and stomach fullness (% body weight) between laboratories.
4. Assess diel changes in prey composition and diet overlap.
5. Evaluate statistical/quantitative techniques for describing diet overlap and dietary preference and develop sample size estimates for the 1995 season.

4. Methods:

Objective 1:

A stratified-random sampling design will be employed to estimate diet overlap and prey electivity among forage fish species. Strata will be established based upon date (May, June, July, August) and habitat type (shallow bay, moderate-slope passage, offshore). Site will be used as the sample unit in the analysis. Samples will be collected between 1500 and 2100 hours from approximately 12 randomly selected sites within each stratum. A randomly selected sample of fifteen individuals (whole fish) will be preserved (10% buffered formaldehyde solution) from each species in each net set. In cases where distinct size classes occur within species, samples will be preserved from each size class. Size related shifts in diet have been noted in several fish species including Pacific cod (*Gadus macrocephalus*, Livingston 1989) and walleye pollock (*Theragra chalcogramma*, Dwyer et al. 1987).

During May and June, the field sampling for this project will be conducted as part of the SEA program (Salmon Growth and Mortality and Salmon Predation components). In nearshore habitats and shallow bays, fish will be collected using tow nets (5 m X 3 m, 1.0 cm stretch mesh in codend), beach seines (40 m long, 0.5 cm stretch mesh), and purse seines (70 m long, 0.5 cm stretch mesh) deployed from a 6 m long aluminum skiff. An approximately 25 m long vessel will provide logistical support to the field crew (see SEA: Salmon Growth and Mortality DPD). In shallow nearshore habitats, fish schools will be located from visual surveys. The Nearshore Fish component of the SEA program will provide data on

fish distribution to the sampling crew. Fish schools will be randomly sampled. An approximately 25 m trawl vessel will sample fish in offshore areas using a 40 m x 28 m mid-water wing trawl equipped with a net sounder (see SEA: Salmon Predator DPD). The cod end of the the trawl will be lined with approximately 2.0 cm stretch-mesh web to retain small specimens. Data from the net sounder will be used to insure that the number of fish caught in each set does not greatly exceed required sample sizes. A smaller mid-water trawl net 5 m x 3 m (cod-end 1.0 cm stretch mesh) will be used to sample small fish in deep water areas.

During July and August, the field sampling for this project will be conducted in cooperation with the NOAA contractor. The sampling gear used in nearshore habitats will be the same as that described above (see SEA: Salmon Growth and Mortality DPD). In offshore habitats, samples will be collected with a mid-water trawl net. Net specifications will be determined by the NOAA contractor.

Quantitative samples of available prey resources will be collected at each site where fish are taken for stomach contents analysis. A ring-net (0.5 m diameter, 100 micron mesh) will be towed vertically from 2 m above the substrate to the surface. Epibenthic prey will be sampled with a pump at five replicate sites near each net-set site. A plexiglass frame (0.6 m x 0.6 m x 1 m) will be placed over the substrate at each sample site, and epibenthic animals removed with the pump. Each sample will be passed through a ring net (100 micron mesh) to retain prey animals. Epibenthic invertebrate samples from the five replicate sites will be combined in a single sample bottle. All samples will be preserved in 10% buffered formaldehyde solution.

Objective 2:

Stomach contents analyses will be conducted after a minimum of 20 days of preservation in 10% formaldehyde solution. Whole fish will be blot dried, weighed to the nearest 0.01 g, and measured to the nearest 0.5 mm. Fish wet weight will be converted to dry weight using literature conversion factors when possible (Volk et al. 1984, Harris 1986). If literature values are not available, wet weight to dry weight conversion factors will be estimated from a sample (n = 100) of individuals from a range of sizes. Total stomach weight including contents and lining will be measured to an accuracy of 0.1 mg. Fish showing evidence of regurgitation will not be included in the analysis. A sample of individuals from a range of sizes (n = 100) will be used to estimate a regression equation relating fish total length to stomach lining weight. Total stomach contents weight will be estimated by subtracting estimated stomach lining weight from total stomach weight. Prey items in the gut will be identified to the lowest possible taxonomic level and enumerated. Prey biomass in each category will be estimated by the product of prey abundance and average prey dry weight (Cooney et al. 1981, J. Landingham pers. comm.). When literature values are not available, mean dry weight will be determined by weighing a sample of intact specimens of each taxon dried at 60°C until weight is stabilized. Diet composition will be expressed as a proportion of total abundance and stomach contents weight. Stomach fullness will be expressed as a proportion

of fish body (dry) weight.

The composition of available prey resources will be estimated from laboratory analyses of ring net and epibenthic pump samples. Zooplankton and epibenthic invertebrates will be identified to the lowest possible taxonomic level. A Hansen-stempel pipette will be used to collect two 1 ml random subsamples from each sample bottle. Animals will be identified and enumerated in each subsample. Total biomass in each taxonomic group will be estimated by the product of average body weight and abundance. Literature values for average body weight of each species or size class will be used when available (Cooney et al. 1981, J. Landingham pers. comm.). When literature values are not available, mean dry weight will be determined by weighing a sample of intact specimens from each taxon dried at 60°C until weight is stabilized. The composition of available prey will be described by pooling the data from epibenthic and zooplankton samples standardized to a 1 m² surface area.

Objective 3:

A subsample of each forage species/size-class from each site ($n \approx 5$ fish) will be sent to each of the two laboratories for analysis of stomach contents. A Paired-t statistic will be used to test for differences between laboratories in the measurement of absolute and relative abundance and biomass of each prey item and in the measurement of stomach fullness. A Multivariate analysis of variance (MANOVA) statistic will be used initially to test for no overall laboratory effect on diet composition of each forage fish. Tests will be conducted at the $P = .05$ significance level.

Objective 4:

Forage fish samples ($n = 15$) will be collected for stomach contents analysis in one habitat type at midnight, 0400, 0800, 1200, 1600, and 2000 hours. A study area will be selected where several species of forage fish are available. Samples will be collected and processed as described under objectives 1 & 2. MANOVA (for biomass data) and a discrete data analysis (for abundance data) will be used to test for temporal changes in diet composition of the forage fish species. A transformation of the data may be required for analysis of variance procedures (Willette 1993). Diet overlap indices (see objective 5) will be used to evaluate diel patterns of diet similarity between pairs of forage fish. If significant diel changes in diet overlap are detected, time of day will be incorporated into the sampling design in future years.

Objective 5:

Several statistical/quantitative methods of assessing diet overlap and prey composition patterns will be investigated. Occurrence rates of prey items in the diet of each forage species will be recorded prior to pooling the data from each net sample. Diet overlap indices such as Morisita's (1959) Measure for abundance

data and Horn's (1966) Index for biomass data will be used to describe the degree of diet overlap between pairs of forage species and to test for differences among habitat types (see Krebs 1989). A Multivariate ordination method (e.g., correspondence analysis) will be used to graphically evaluate diet similarity patterns and prey resource composition and to test for differences in diet and prey composition among habitat types (see Ludwig and Reynolds 1988, Digby and Kempton 1987). Ivlev's (1961) electivity index and Manly's alpha (Manly et al. 1972) will be used to measure dietary preferences of each forage species. Preference for each available prey item will be compared among forage species and habitat types. MANOVA methods will be used to statistically assess prey and diet composition and dietary preference (e.g., Manly 1986, Johnson and Wichern 1988). If necessary, the data will be transformed to meet the assumption of residual normality. Results from these analyses will be used to help develop a broad-scale sampling design for the 1995 season. Variance estimates of diet overlap and prey composition will be used to select within-stratum sample sizes to meet desired precision requirements (Cochran 1977).

5. Location:

The field sampling component of this study will be conducted in PWS. Laboratory analyses will be conducted at the NMFS Auke Bay Laboratory and University of Alaska Fairbanks.

6. Technical Support:

Technical support regarding laboratory prey resource and stomach contents analyses will be provided by staff at the NMFS Auke Bay Laboratory and University of Alaska Fairbanks.

7. Contracts:

The University of Alaska Fairbanks will provide laboratory stomach contents analysis through a Restricted Services Agreement.

D. SCHEDULES

The field season for this project will be from April to August of each year. Laboratory and data analysis will be conducted during the remainder of the year (Table 1).

Table 1: Schedule of project activities related to 1994 field season.

Time Period	Activity
<u>Field Studies</u>	
May 15 - May 26	Field Sampling in SEA program
June 15 - June 26	Field Sampling in SEA program
July 15 - July 26	Field Sampling with NOAA contractor
August 15 - August 26	Field Sampling with NOAA contractor
<u>Laboratory & Data Analyses</u>	
June 1 - December 31, 1994	Conduct stomach contents analysis
January 1, 1995 - March 31, 1995	Analyze data and prepare annual report.
April 1, 1995	Submit annual project report

E. EXISTING AGENCY PROGRAM

F. ENVIRONMENTAL COMPLIANCE/PERMIT/COORDINATION STATUS

This project will qualify for an exclusion from the requirements of the National Environmental Policy Act. The project will not cause a significant environmental impact.

G. PERFORMANCE MONITORING

An annual report detailing the results from the previous year's investigations will be submitted by April 1 of each year.

H. COORDINATION OF INTEGRATED RESEARCH EFFORT

This project will be highly integrated with several components of the SEA Program. The Physical Oceanography, Nearshore Fish, Zooplankton, and Phytoplankton components of SEA will collect data relevant to forage fish distribution and production. Within the Physical Oceanography component, conductivity-temperature-depth (CTD) profilers and Acoustic Doppler Current Profilers (ADCP) will be deployed from a mid-water trawl vessel. Within the Nearshore Fish component, hydroacoustic data will be obtained in offshore habitats from a mid-water trawl vessel and in nearshore habitats from small hydroacoustic survey boats. Within the Zooplankton and Phytoplankton components, zooplankton and water samples will be collected using nets and water bottles. The Salmon Growth and Salmon Predation components of SEA will collect forage fish samples for later stomach contents analysis in offshore and nearshore habitats using mid-water trawls, and beach and purse seines. Age-weight-length data will be collected from the forage fish to accompany hydroacoustic data. Salmon Predation will provide a research platform for one marine bird observer in cooperation with the USFWS component of Forage Fish. An additional marine bird observer will be accommodated on each of the seine vessels, if possible. All data collected as part of SEA will be provided to the Information and Modeling component for use in development and implementation of ecosystem models.

I. PUBLIC PROCESS

J. PERSONNEL QUALIFICATIONS

Mark Willette
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. Conduct various fisheries enhancement and evaluation projects in PWS including juvenile salmon growth studies, lake stocking, limnological investigations of sockeye salmon producing lakes, and quality control of coded-wire tagging at private hatcheries. Conduct fisheries oceanographic studies in PWS in cooperation with private hatcheries and University of Alaska investigators. Chairman of

PWS Regional Planning Team. Principal Investigator: 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. 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.

EDUCATION:

1985 Master of Science, Fisheries Oceanography, University of Alaska Fairbanks.

1983 Bachelor of Science, Fisheries Science, University of Alaska Fairbanks.

Stan R. Carlson
Alaska Department of Fish and Game
Commercial Fisheries Management and Development Division
34828 Kalifornsky Beach Rd, Suite B
Soldotna, Alaska 99669
(907)262-9368

EMPLOYMENT:

January 1993- present: Biometrician for the Alaska Department of Fish and Game, Limnology Section, Commercial Fisheries Management and Development Division, Soldotna, Alaska. Supervised by Dr. Dana Schmidt. Conduct statistical data analyses to evaluate factors that affect dynamics of the biota in lake ecosystems. Design limnological experiments and determine methods to estimate zooplankton and salmon abundance. Develop and approve methods to estimate hatchery contributions to the fishery. Develop, review, and conduct statistical analyses for projects related to the impact of oil on commercial fishery species. Provide biometrical consulting to area and

regional biologists and statewide limnologists.

November 1991 - January 1993: Mathematical Statistician for the National Marine Fisheries Service, Auke Bay Laboratory, Juneau, Alaska. Supervised by Mr. Steven Ignell. Conduct statistical studies on community attributes of pelagic fauna in the north Pacific Ocean. Provide biometrical consulting, technical editing, and collaborative input on projects such as salmon bycatch and climate change studies.

January 1989 - May 1991: Statistics Teacher, Experimental Statistics Department, New Mexico State University, Las Cruces. Supervised by Dr. Michael Ames. Instruct laboratory courses in statistics for undergraduate science majors.

May - August 1990: Research Specialist (statistician), Department of Entomology, Plant Pathology, and Weed Science, New Mexico State University. Dr. Ellis Huddleston, Supervisor. Provide statistical modeling, analysis, and design of experiments related to agricultural field studies and pest management programs.

May - December 1988: Field Biologist, Biology Department, New Mexico State University. Supervised by Mr. Roger Skaggs. Conduct field population surveys and habitat analyses of night birds in Lincoln National Forest, New Mexico. Collect field data, supervise field personnel, and maintain data records. Develop operational strategies and conduct follow-up statistical estimation procedures.

August 1985 - June 1988: Graduate Assistant, Biology Department, New Mexico State University. Supervised by Dr. Ralph Raitt and Dr. Walt Whitford. Teach undergraduate biology and zoology laboratory courses. Collect data and maintain field ecology experiments for ecological research programs. Develop and conduct original field research on desert insect ecology.

June 1983 - May 1985: Research Specialist, Gordon Environmental Studies Laboratory, University of Montana, Missoula. Supervised by Dr. Philip Tourangeau. Manage data, conduct quality assurance/control procedures, and perform statistical analyses for environmental science projects. Aid in the design and implementation of field research, primarily in the area of pollution biomonitoring.

EDUCATION:

1991 Master of Science, Experimental Statistics, New Mexico State University.

1988 Master of Science, Biology (ecology), New Mexico State University.

K. BUDGET

Table 2: Budget summary for the Salmon Predation component of the SEA program in FY94, FY95, and FY96 and beyond. Budgets for FY95 and beyond may change as information from the first year of study is applied to refine the methodology.

Line Item	FY94	FY95	FY96 and beyond
Personnel	23.7	46.5	46.5
Travel	0.0	1.2	1.2
Contractual	41.6	103.6	103.6
Supplies	1.4	2.8	2.8
Equipment	0.0	0.0	0.0
Total	66.7	154.1	154.1
Indirect Costs	8.7	14.2	14.2
Grand Total	75.4	168.3	168.3

L. REFERENCES

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- Cooney, R.T., D. Urquhart, and D. Barnard. 1981. The behavior, feeding biology, and growth of hatchery released pink and chum salmon fry in Prince William Sound, Alaska. University of Alaska, Sea Grant Rep. 81-5. 114p.
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- Krebs, C.J. 1989. Ecological Methodology. Harper and Row, New York.
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- Livingston, P.A. 1989. Interannual trends in Pacific cod, *Gadus macrocephalus*, predation on three commercially important crab species in the eastern Bering Sea. Fish. Bull. 87: 807-827.
- Ludwig, J.A. and J.F. Reynolds. 1988. Statistical Ecology: a primer on methods and computing. John Wiley and Sons, Inc., New York.
- Manly, B.F.J., P. Miller, L.M. Cook. 1972. Analysis of a selective predation experiment. Amer. Nat. 106: 719-736.

Manly, B.F.J. 1986. Multivariate statistical methods: a primer. Chapman and Hall, London and New York.

Morisita, M. 1959. Measuring of interspecific association and similarity between communities. Mem. Fac. Sci. Kyushu Univ. Ser. E(Biol.) 3: 65-80.

Volk, E.C., R.C. Wissmar, C.A. Simenstad, and D.M. Eggers. 1984. Relationship between otolith microstructure and the growth of juvenile chum salmon (*Oncorhynchus keta*) under different prey rations. Can. J. Fish. Aquat. Sci. 41: 126-133.

Willette, T.M. 1993. 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, February 1993, Anchorage, Alaska (in press).

EXXON VALDEZ TRUSTEE COUNCIL
1994 Federal Fiscal Year Project Budget
October 1, 1993 - September 30, 1994

Project Description: Forage Fish Influence on Injured Species - This project will determine temporal and spatial distribution, abundance, and availability of important prey species (e.g. herring, pollack, sand lance, caplin, euphausiids, copepods) in PWS; determine how important biotic and abiotic factors affect both short- and long-term distribution and abundance of prey species; and determine how predator distribution, abundance, and foraging strategy coincide with forage fish distribution and abundance.

Budget Category:	1993 Project No. Authorized FFY 93	'93 Report/ '94 Interim* FFY 94	Remaining Cost** FFY 94	Total FFY 94	FFY 95	Comment
Personnel	\$0.0	\$0.0	\$130.1	\$130.1	\$96.5	
Travel	\$0.0	\$0.0	\$14.0	\$14.0	\$13.2	
Contractual	\$0.0	\$0.0	\$391.6	\$391.6	\$603.6	
Commodities	\$0.0	\$0.0	\$6.7	\$6.7	\$2.8	
Equipment	\$0.0	\$0.0	\$0.0	\$0.0	\$0.0	
Capital Outlay	\$0.0	\$0.0	\$0.0	\$0.0	\$0.0	
Subtotal	\$0.0	\$0.0	\$542.4	\$542.4	\$716.1	
General Administration	\$0.0	\$0.0	\$44.2	\$44.2	\$44.2	
Project Total	\$0.0	\$0.0	\$586.6	\$586.6	\$760.3	
Full-time Equivalents (FTE)	0.0	0.0	2.3	2.3	1.4	
Dollar amounts are shown in thousands of dollars.						
Budget Year Proposed Personnel:		Reprt/Intrm	Reprt/Intrm	Remaining	Remaining	
Position Description		Months	Cost	Months	Cost	
See Individual 3A Forms for Personnel Details						
Personnel Total		0.0	\$0.0	0.0	\$0.0	
NEPA Cost:						\$0.0
*Oct 1, 1993 - Jan 31, 1994						
**Feb 1, 1994 - Sep 30, 1994						

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Project Number: 94163
 Project Title: Forage Fish Influence on Injured Species
 Agency: National Oceanic & Atmospheric Admin.

FORM 2A
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EXXON VALDEZ TRUSTEE COUNCIL
1994 Federal Fiscal Year Project Budget
October 1, 1993 - September 30, 1994

Project Description: Forage Fish Influence on Injured Species - This project will determine temporal and spatial distribution, abundance, and availability of important prey species (e.g. herring, pollack, sand lance, caplin, euphausiids, copepods) in PWS; determine how important biotic and abiotic factors affect both short- and long-term distribution and abundance of prey species; and determine how predator distribution, abundance, and foraging strategy coincide with forage fish distribution and abundance.

Budget Category:	1993 Project No. Authorized FFY 93	'93 Report/ '94 Interim* FFY 94	Remaining Cost** FFY 94	Total FFY 94	FFY 95	Comment
Personnel	\$0.0	\$0.0	\$66.0	\$66.0	\$50.0	
Travel	\$0.0	\$0.0	\$10.0	\$10.0	\$12.0	
Contractual	\$0.0	\$0.0	\$350.0	\$350.0	\$500.0	
Commodities	\$0.0	\$0.0	\$0.0	\$0.0	\$0.0	
Equipment	\$0.0	\$0.0	\$0.0	\$0.0	\$0.0	
Capital Outlay	\$0.0	\$0.0	\$0.0	\$0.0	\$0.0	
Subtotal	\$0.0	\$0.0	\$426.0	\$426.0	\$562.0	
General Administration	\$0.0	\$0.0	\$29.4	\$29.4	\$30.0	
Project Total	\$0.0	\$0.0	\$455.4	\$455.4	\$592.0	
Full-time Equivalents (FTE)	0.0	0.0	0.8	0.8	0.6	
Dollar amounts are shown in thousands of dollars.						
Budget Year Proposed Personnel:		Reprt/Intrm Months	Reprt/Intrm Cost	Remaining Months	Remaining Cost	
Position Description						
Program Manager GS-12		0.0	\$0.0	1.0	\$6.0	
Project Leader GS-13		0.0	\$0.0	8.0	\$60.0	
Personnel Total		0.0	\$0.0	9.0	\$66.0	
NEPA Cost:						\$0.0
*Oct 1, 1993 - Jan 31, 1994						
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Project Number: 94163
 Project Title: Forage Fish Influence on Injured Species
 Sub-Project: Hydroacoustic Surveys
 Agency: National Oceanic & Atmospheric Admin.

FORM 3A
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 DETAIL

EXXON VALDEZ TRUSTEE COUNCIL
 1994 Federal Fiscal Year Project Budget
 October 1, 1993 - September 30, 1994

Travel:	Reprt/Intrm	Remaining
Travel To Anchorage, Juneau and Seattle - 5 trips at \$1K/trip	\$0.0	\$5.0
Travel to Cordova and Prince William Sound	\$0.0	\$5.0
Travel Total	\$0.0	\$10.0
Contractual:		
Contractor to design and conduct reconnaissance surveys for forage fish in the summer of 1994.	\$0.0	\$350.0
Contractual Total	\$0.0	\$350.0

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Project Number: 94163
 Project Title: Forage Fish Influence on Injured Species
 Sub-Project:
 Agency: National Oceanic & Atmospheric Admin.

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 DEVELOPMENT

EXXON VALDEZ TRUSTEE COUNCIL
1994 Federal Fiscal Year Project Budget
October 1, 1993 - September 30, 1994

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Project Number: 94163
Project Title: Forage Fish Influence on Injured Species
Sub-Project:
Agency: National Oceanic & Atmospheric Admin.

FORM 3B
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DETAIL

EXXON VALDEZ TRUSTEE COUNCIL
1994 Federal Fiscal Year Project Budget
October 1, 1993 - September 30, 1994

Project Description: Forage Fish Influence on Injured Species - The ADF&G portion of this project will estimate the degree of diet overlap among forage fish species including juvenile salmon. This project will collect and analyze stomach samples from forage fish in nearshore habitats and sample available prey resources. Diet overlap and prey selectivity indices will be calculated and multivariate analysis of variance techniques applied to examine resource partitioning among forage fish.

Budget Category:	1993 Project No. Authorized FFY 93	'93 Report/ '94 Interim* FFY 94	Remaining Cost** FFY 94	Total FFY 94	FFY 95	Comment
Personnel	\$0.0	\$0.0	\$23.7	\$23.7	\$46.5	
Travel	\$0.0	\$0.0	\$0.0	\$0.0	\$1.2	
Contractual	\$0.0	\$0.0	\$41.6	\$41.6	\$103.6	
Commodities	\$0.0	\$0.0	\$1.4	\$1.4	\$2.8	
Equipment	\$0.0	\$0.0	\$0.0	\$0.0	\$0.0	
Capital Outlay	\$0.0	\$0.0	\$0.0	\$0.0	\$0.0	
Subtotal	\$0.0	\$0.0	\$66.7	\$66.7	\$154.1	
General Administration	\$0.0	\$0.0	\$8.7	\$8.7	\$14.2	
Project Total	\$0.0	\$0.0	\$75.4	\$75.4	\$168.3	
Full-time Equivalents (FTE)	0.0	0.0	0.5	0.5	0.8	
Dollar amounts are shown in thousands of dollars.						
Budget Year Proposed Personnel: Position Description	Reprt/Intrm Months	Reprt/Intrm Cost	Remaining Months	Remaining Cost		
Fishery Biologist I	0.0	\$0.0	1.8	\$7.1		
Fishery Biologist I	0.0	\$0.0	1.8	\$7.1		
Fish and Wildlife Technician II	0.0	\$0.0	2.0	\$6.1		
Program Manager	0.0	\$0.0	0.5	\$3.4		
Personnel Total	0.0	\$0.0	6.0	\$23.7		
					NEPA Cost:	\$0.0
					*Oct 1, 1993 - Jan 31, 1994	
					**Feb 1, 1994 - Sep 30, 1994	

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Project Number: 94163
 Project Title: Forage Fish Influence on Injured Species
 Sub-Project:
 Agency: AK Dept. of Fish and Game

FORM 3A
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 DATA

EXXON VALDEZ TRUSTEE COUNCIL
1994 Federal Fiscal Year Project Budget
October 1, 1993 - September 30, 1994

Travel:	Reprt/Intrm	Remaining
Travel Total	\$0.0	\$0.0
Contractual:		
Air charter to sampling sites (9 hours @ \$400/hour)	\$0.0	\$3.6
Contract for laboratory stomach sample processing	\$0.0	\$38.0
Contractual Total	\$0.0	\$41.6

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Project Number: 94163
Project Title: Forage Fish Influence on Injured Species
Sub-Project:
Agency: AK Dept. of Fish & Game

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EXXON VALDEZ TRUSTEE COUNCIL
 1994 Federal Fiscal Year Project Budget
 October 1, 1993 - September 30, 1994

Commodities:		Reprt/Intrm	Remaining
Lab supplies (preservatives, fixatives, stains)		\$0.0	\$0.4
Field sampling supplies (containers, formalin, measuring boards)		\$0.0	\$1.0
Commodities Total		\$0.0	\$1.4
Equipment:			
Equipment Total		\$0.0	\$0.0

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Project Number: 94163
 Project Title: Forage Fish Influence on Injured Species
 Sub-Project:
 Agency: AK Dept. of Fish & Game

FORM 3B
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 PROJECT
 DETAIL

EXXON VALDEZ TRUSTEE COUNCIL
1994 Federal Fiscal Year Project Budget
October 1, 1993 - September 30, 1994

Project Description: Forage Fish Influence on Injured Species - This project will determine temporal and spatial distribution, abundance, and availability of important prey species (e.g. herring, pollack, sand lance, caplin, euphausiids, copepods) in PWS; determine how important biotic and abiotic factors affect both short- and long-term distribution and abundance of prey species; and determine how predator distribution, abundance, and foraging strategy coincide with forage fish distribution and abundance.

Budget Category:	1993 Project No. Authorized FFY 93	'93 Report/ '94 Interim* FFY 94	Remaining Cost** FFY 94	Total FFY 94	FFY 95	Comment
Personnel	\$0.0	\$0.0	\$40.4	\$40.4	\$0.0	
Travel	\$0.0	\$0.0	\$4.0	\$4.0	\$0.0	
Contractual	\$0.0	\$0.0	\$0.0	\$0.0	\$0.0	
Commodities	\$0.0	\$0.0	\$5.3	\$5.3	\$0.0	
Equipment	\$0.0	\$0.0	\$0.0	\$0.0	\$0.0	
Capital Outlay	\$0.0	\$0.0	\$0.0	\$0.0	\$0.0	
Subtotal	\$0.0	\$0.0	\$49.7	\$49.7	\$0.0	
General Administration	\$0.0	\$0.0	\$6.1	\$6.1	\$0.0	
Project Total	\$0.0	\$0.0	\$55.8	\$55.8	\$0.0	
Full-time Equivalents (FTE)	0.0	0.0	1.0	1.0	0.0	
Dollar amounts are shown in thousands of dollars.						
Budget Year Proposed Personnel:						
Position Description		Reprt/Intrm Months	Reprt/Intrm Cost	Remaining Months	Remaining Cost	
Principal Investigator, Biologist		0.0	\$0.0	6.0	\$24.4	
Supervisory Biologist		0.0	\$0.0	1.0	\$5.0	
Biotechnician		0.0	\$0.0	5.0	\$11.0	
Personnel Total		0.0	\$0.0	12.0	\$40.4	
					NEPA Cost:	\$0.0
					*Oct 1, 1993 - Jan 31, 1994	
					**Feb 1, 1994 - Sep 30, 1994	

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Project Number: 94163
 Project Title: Forage Fish Influence on Injured Species
 Sub-Project: Predator Surveys
 Agency: Dept. of Interior, Fish & Wildlife Service

FORM 3A
SUB-
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DETAIL

EXXON VALDEZ TRUSTEE COUNCIL
 1994 Federal Fiscal Year Project Budget
 October 1, 1993 - September 30, 1994

Travel:	Reprt/Intrm	Remaining
Anchorage to Prince William Sound (6 trips - transportation \$200/trip + 10 days per diem @ \$3/day)	\$0.0	\$1.6
Anchorage to Juneau (4 trips @ \$600/trip)	\$0.0	\$2.4
Travel Total	\$0.0	\$4.0
Contractual:		
Contractual Total	\$0.0	\$0.0

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Project Number: 94163
 Project Title: Forage Fish Influence on Injured Species
 Sub-Project: Predator Surveys
 Agency: Dept. of Interior & Wildlife Service

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 DETAIL

EXXON VALDEZ TRUSTEE COUNCIL
 1994 Federal Fiscal Year Project Budget
 October 1, 1993 - September 30, 1994

Commodities:		Reprt/Intrm	Remaining
Boat fuel (30 days x \$100/day)		\$0.0	\$3.0
Boat oil (30 days x \$10/day)		\$0.0	\$0.3
Boat preparation and maintenance		\$0.0	\$2.0
Commodities Total		\$0.0	\$5.3
Equipment:			
Equipment Total		\$0.0	\$0.0

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Project Number: 94163
 Project Title: Forage Fish Influence on Injured Species
 Sub-Project: Predator Surveys
 Agency: Dept. of Interior, Fish & Wildlife Service

FORM 3B
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PROJECT
DETAIL

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94165

THIS PROJECT IS ON HOLD

**PROJECT 94165
HERRING: GENETIC STOCK IDENTIFICATION
IN PRINCE WILLIAM SOUND**

**EXXON VALDEZ TRUSTEE COUNCIL
FY 94 DETAILED PROJECT DESCRIPTION**

Project title: Herring Spawn Deposition and Reproductive Impairment

Project ID number: 94166

Project type: Research / Monitoring

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Cooperating agencies: NOAA

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Project Leader: Linda Brannan for John Wilcock 3/4/94
John Wilcock Date

Project Manager: Joseph R. Sullivan 3/4/94
Joe Sullivan Date

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EXXON VALDEZ OIL SPILL
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INTRODUCTION

The Exxon Valdez oil spill coincided with the spring migration of Pacific herring *Clupea pallasii* to spawning grounds in 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. Research continued through 1992 with contributions from both state general funds and the Trustee Council. 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 (Reid 1971). 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 is 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. Consequently, a pilot experiment to measure the ability of herring from this age class to produce viable offspring was conducted in 1992. Hatching success of eggs collected from fish spawning in previously oiled areas was less than half that of eggs collected from fish spawning in pristine areas. There were approximately twice as many abnormal larvae from fish spawning in the previously oiled areas as well.

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. Of the four commercial herring fisheries which normally occur each spring, guideline harvests were attained for only one gear type (gillnet sac roe). Purse seine sac roe fishermen, who typically average about 66% of the annual harvest, did not realize any harvest due to low commercial quality related to fish size and low abundance of herring. A preliminary pathology study implicated viral hemorrhagic septicemia (VHS) as a potential source of mortality and stress. It is not clear whether the Exxon Valdez oil spill may be implicated, although numerous studies have indicated that previous exposure to toxins can reduce immunity to disease.

This project provides a direct measure of herring abundance that is vital to monitoring recovery of the injured PWS herring population and also information about reproductive biology that is necessary for improving the interpretation of earlier damage assessment

results. Project results will improve our understanding of long term damage and can also be used in setting commercial harvest strategies, thereby contributing to the recovery process for PWS herring populations.

The cost of this project is reasonable considering the economic value of the commercial fisheries as well as the important contribution that herring of all life stages make to the PWS ecosystem. The ex-vessel value of the herring fisheries in 1992 was \$12.0 with an average annual value of \$8.3 million. In 1993, the exvessel value dropped to \$2.0 million because only one third of the expected harvestable population returned and was made up largely of small fish with a low market value. This project will enable resource managers to better understand herring population dynamics to improve the recovery process. In addition, it will aid local resource users to make appropriate pre-season plans based on accurate and precise herring return projections.

PROJECT DESCRIPTION

This project will be conducted in three parts. ADF&G will be performing the two field components which constitute the continuation of both the herring spawn deposition survey and egg loss study in PWS. NOAA will be performing the laboratory component of this project which constitutes the reproductive impairment study. This detailed plan covers only the ADF&G component of spawn deposition survey and egg loss study. A separate plan will be submitted by NOAA to cover reproductive impairment (Mark Carls, NMFS, Auke Bay Lab).

Spawn deposition surveyors will collect basic biological information about the abundance and distribution of herring embryos in PWS. This information will be used to estimate adult spawning biomass, a crucial element to forecasting adult herring returns and to monitoring resource recovery. Annual forecasts of returns form the basis of herring fishery management in PWS. Data collected during spawn deposition surveys will include location, substrate type, vegetation type, egg density, gradient, and nearby sea surface temperature of herring spawning beds. These data will be incorporated into an existing database containing over 22,000 data points collected during studies in 1983, 1984, and 1988-1992. New and existing data will be used to develop a classification system for spawning habitat types. Habitat use and spawner/egg density and distribution data collected during this project are needed to develop herring embryo survival models as outlined in the Natal Habitat Program (NHP) of the SEA plan (Project 94320).

The egg loss study will provide estimates of herring embryos physically removed from spawning areas by predation and wave action. This information is useful for two reasons (1) to account for the loss of eggs between the time of spawning and the time of spawn deposition surveys to improve the accuracy of spawning biomass estimates, and (2) to estimate total embryo survival. This second reason is vital to implementation of the NHP component of the SEA proposal. Determining the relationship between egg loss and egg density, habitat type, predation levels, and meteorological conditions is crucial to the development of a valid herring embryo survival model from which to estimate annual larval herring production in PWS. Ultimately, a better understanding of larval production dynamics

will improve our understanding of processes affecting herring recruitment.

Resources and/or Associated Services:

Pacific herring *Clupea pallasii* are a major resource in Prince William Sound (PWS) from both commercial and ecological perspectives. Five commercial herring fisheries in PWS have an average annual combined ex-vessel value of \$8.3 million. 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, rockfish, and other fish. 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 Chenega and Tatitlek.

The two primary goals of the proposed project are to improve the accuracy of fisheries management of the PWS herring resource and to begin implementation of the NHP component of the SEA plan. Accurate and precise estimation of herring abundance and the processes affecting herring production is crucial to the improvement of management accuracy as well as our understanding of the PWS ecosystem. Improved accuracy 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 and additional damage to the resource or of under harvest and economic loss to local communities. Because commercial and subsistence herring harvests represent substantial contributions to local economies, intensive management is expected to benefit all communities in PWS.

Relation to Other Damage Assessment/Restoration Work:

This project is in part a continuation of EVOS Natural Resource Damage Assessment Fish/Shellfish Study No. 11, Injury to PWS Herring, which was conducted from 1989 through 1991. A close-out study was conducted in 1992. No field work was conducted in 1993 and funding for all herring research ended in July. The apparent crash of the PWS adult herring population in 1993 has renewed interest in pursuing herring research from a restoration standpoint.

Previous egg loss studies to determine the removal of eggs due to predation and wave action were conducted in PWS in 1990 and 1991. In addition, embryo mortality studies to estimate the survival of remaining eggs not removed were conducted in 1989-1991. Results from these investigations will be incorporated with results from the current study to begin building an embryo survival model.

This project will provide information required by the NHP component of the SEA plan (Project 94320). One NHP study included in the SEA plan and slated for implementation in 1994, Assessment of Avian Predation on Herring Spawn, will be conducted by the Copper River Delta Institute, USFS, Cordova District. The avian predation study will be carried out

cooperatively with the spawn deposition project and will include synoptic data collection.

Other research programs from the 1994 work plan that will require close cooperation with the spawn deposition project for sharing of data, personnel, or other resources include: (1) Project 94163, Forage Fish Influence on Injured Species; (2) Project 94165, Herring Genetic Stock Identification in PWS; and (3) the Reproductive Impairment portion of the current project which will be conducted by NOAA (Mark Carls, Auke Bay, Project Leader).

Objectives:

Specific objectives of this project for the 1994 work plan include:

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.
2. Quantify egg loss rates (the proportion of eggs removed through time) from spawning areas due to wave action, predation, dessication, or fungal infections between the time of egg deposition (spawning) and the time of hatching. Quantify egg loss by habitat type and egg density.
3. Incorporate egg loss and egg survival estimates with results from previous studies and revise the models as necessary.

Additional objectives that should be included in the 1995 work plan as a continuation of this project are:

4. Define herring spawning habitat types by similarities in temperature, salinity, depth, gradient, substrate, vegetation, and exposure to wave action. Characterize and map habitat utilized for spawning. Estimate the abundance and distribution of adult herring and eggs by habitat type. Test a model of the relationship of spawn timing, spawner density and abundance to egg distribution and density.
5. Incorporate egg loss and survival data with physical oceanographic and meteorological data to formulate and test a model of the relationship of meteorological conditions to wave height and egg desiccation.
6. Test a model of the relationship between predation, wave action, desiccation, fungal infections, habitat type, and egg density.
7. Test a model relating sound-wide embryo survival to habitat type, egg density, and meteorological conditions.

Methods:

Spawn Deposition Survey and Biomass Estimation

Spawn deposition survey design was modified in 1989 for NRDA studies to more accurately assess the PWS herring stock's response to the 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 estimation based on spawn deposition surveys consisted 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 follows closely the two-stage sampling design of similar surveys in British Columbia (Schwiebert et al. 1985) and Southeast Alaska (Blankenbeckler and Larson 1982, 1987). Surveys will use random sampling for the first stage (transects) and systematic sampling for the second stage (quadrats within transects). Random sampling for the second stage is not feasible because of underwater logistical constraints (Schwiebert et al. 1985). In addition, surveys will be stratified by area to account for geographic differences and the potential for discrete herring stocks. Areas surveyed 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 will be used to estimate total number of eggs deposited in each of the five areas. 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 will be needed to ensure that estimated biomass would be within 25% of the true biomass 95% of the time. There are 3,163 potential transects per kilometer based on the size of the sampling quadrat and so 100 km of herring spawn would require 110 transects to maintain the accuracy goal. Confidence intervals will be calculated assuming a normal distribution of total egg estimates.

Spawn Deposition Survey Sampling Procedure. The general location of spawning activity will be determined from milt observed during scheduled aerial surveys which are part of the 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 estimated from aerial survey maps will be corrected by skiff and diver surveys at the time of the dive survey. 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 at low tide.

Each shoreline area containing herring spawn will be divided into the smallest resolvable segments on the map scale (approximately 0.18 km) and the segments numbered. Total potential transects will be calculated from the total shoreline km of observed spawn and a minimum of 0.035% 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. Each team will work a morning and afternoon shift daily. Based on information from previous PWS surveys, two diving teams can generally complete 6 to 12 transects each day 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. However, if new divers are included in the survey, training may require about one additional week.

The exact location for each survey transect will be fixed as the dive skiff approaches the shore 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.

Using a sampling quadrat consisting of a 0.1 m² frame constructed of PVC pipe with a depth gauge and compass attached, the first quadrat location will be randomly selected within the first 5 meters of spawn. Succeeding quadrat locations will be systematically spaced every 5 meters along the compass course until the apparent end of the spawn is found. Within each quadrat, the lead diver will estimate the number of eggs in units of thousands (K) in the quadrat, communicating the numbers through hand signals to the second diver who will record the estimate. Data will be recorded using a large weighted carpenter's pencil and data forms printed on water-proof plastic paper attached to a PVC clipboard. Vegetation type, percent cover, substrate, and depth will also be recorded for each quadrat. 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 estimate the number of eggs on removable vegetation in each calibration

quadrat independently. All egg-containing vegetation within the quadrat will be removed and placed in numbered mesh bags. The number of loose and attached eggs left after the removal process 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). Currently an ADF&G biometrician is evaluating these proposed sample sizes using historic diver calibration data to ensure that sufficient sampling is planned. 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. Upon completing a dive shift, calibration samples will be preserved in Gilson's solution and carefully 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) and for the 1989 through 1992 surveys (Biggs et al. *in press*). The biomass estimator will be

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

where

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

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

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

where

Var(B') = an unbiased estimate of the variance of B', and
 Var(T) = an unbiased estimate of the variance of T.

Total Number of Eggs (T). The total number of eggs deposited in an area will be estimated from a two-stage sampling program with random sampling at the primary stage, followed by systematic sampling at the secondary stage, using a sampling design similar to that described by Schwiebert et al. (1985). To compute variances based on systematic second stage samples, it will be assumed that eggs will be randomly distributed in spawning beds with respect to the 0.1 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

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

where

- L = the shoreline length of the spawn-containing stratum in meters,
- N = $L/0.1^{0.5}$ = the total number of possible transects,
- $0.1^{0.5}$ = 0.3162 m = width of transect strip,
- \hat{y} = 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

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

where

$$\hat{y}_i = M_i \bar{y}_i$$

and

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

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

$$\bar{y}_i = \frac{\sum_{j=1}^{m_i} y_{ij}}{m_i}, \quad (5)$$

where

- j = quadrat number within transect i ,
- m_i = number of quadrats actually sampled in transect i , and
- y_{ij} = adjusted diver-estimated egg count (in thousands of eggs) from the diver calibration model for quadrat 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 (quadrats), and the variance term for the third stage comes from the regression model used in the diver calibration samples. Therefore the estimated variance of T , conditioned on R , is

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

where

$$s_1^2 = \frac{\sum_{i=1}^n (\bar{y}_i - \bar{y})^2}{n-1} = \quad (7)$$

variance among transects,

$$s_2^2 = \sum_{i=1}^n M_i^2 \sum_{j=1}^{m_i} \frac{(y_{ij} - \bar{y}_i)^2}{n(m_i-1)} = \quad (8)$$

variance among quadrats,

$$s_3^2 = \sum_{i=1}^n \sum_{j=1}^{m_i} \text{Var}(y_{ij}) = \quad (9)$$

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

$$f_1 = \frac{n}{N} = \quad (10)$$

proportion of possible transects sampled, and

$$f_2 = \frac{m_i}{M_i} = \quad (11)$$

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

Diver Calibration. Diver observations of vegetation species will be aggregated into four vegetation categories based on structural and phylogenetic similarities of plants in the quadrat: eelgrass, fucus, hair kelp, and large brown kelp (Becker and Biggs 1992). Diver estimates of egg numbers will be proportional to laboratory-enumerated counts, but systematic biases in the diver estimates will be accounted for by vegetation type and density (Biggs and Funk 1988). Within a year, individual diver effects were not significant for survey divers with at least three years experience, although there were differences among years. Because the experience level of all divers is not guaranteed to be greater than three years in 1994 the diver effect will be included in the model. The basic form of the model currently used to account for biases in diver observations is:

$$Y_{ijk} = e^{\alpha} e^{D_j} e^{V_k} X_{ijk}^{\beta_{jk}} e^{\epsilon_{ijk}}, \quad (12)$$

where

- α = a constant,
- D_j = parameters representing the effect of the j^{th} diver,
- V_k = parameters representing the effect of the k^{th} vegetation type,
- β_{jk} = parameters controlling the functional form of the relationship between the diver estimate and laboratory-enumerated egg count for diver j in vegetation type k ,
- Y_{ijk} = the i^{th} laboratory egg count in the vegetation-diver stratum jk ,
- X_{ijk} = the i^{th} diver estimate in vegetation-diver stratum jk , and
- ϵ_{ijk} = a normally distributed random variable with mean 0 and variance σ^2 .

A multiplicative-effect model is the current choice of models because relative estimation errors were expected to change with egg density. The distribution of laboratory-enumerated egg counts for a given diver and vegetation estimate were positively skewed in the 1988 survey (Biggs and Funk 1988), so that the logarithmic transformation used to estimate the parameters of the multiplicative-effect model also stabilized the variance and corrected the skewness of the egg density estimates. After a logarithmic transformation equation 12 becomes

$$\log_e(Y_{ijk}) = \alpha + D_j + V_k + \beta_{jk} \log_e(X_{ijk}) + \epsilon_{ijk} \quad (13)$$

where

β_{jk} = the slope of the relationship between the logarithm of the diver estimate and the logarithm of the laboratory-enumerated egg count.

In logarithmic form, the model comprises a linear analysis of covariance problem with two factor effects (vegetation and diver) and 1 covariate (diver-estimated egg number). The SAS (1987) procedure for general linear models is currently used to obtain least squares estimates of parameters and evaluate variance components. In addition to the two factor effects and one covariate, terms for diver-vegetation group interactions, density-vegetation group interactions and density-diver interactions is currently considered in the analysis of covariance. Three-way and higher level interaction effects is not considered because we wish to derive a simple model with a relatively small number of parameters. A backward stepwise procedure is used to determine subsets of the six effects that explained the maximum amount of variability in the data with the smallest number of parameters. During the backward stepwise procedure, effects are included or eliminated from the model based on the probability level of F ratios for partial sums of squares.

Translation of predicted values from the logarithmic model, equation 13, back to the original scale, equation 12, requires a correction for bias. The bias in the predicted value of Y_{ijk} is $\exp(\sigma^2/2)$ when the true variance of $\text{Log}(Y_{ijk})$, σ^2 , is known. Laurent (1963) gave an exact expression for the bias correction that incorporated additional terms when σ^2 will be estimated from a sample. For the diver calibration data, the bias encountered from using an estimate of σ^2 was found to be less than 0.05% (Biggs and Funk 1988). Expected values of Y_{ijk} were therefore estimated from

$$E(Y_{ijk}) = e^{\alpha} e^{\hat{D}_j} e^{\hat{V}_k} X_{ijk}^{\hat{\beta}_{jk}} e^{\frac{s^2}{2}}, \quad (14)$$

where

s^2 = the mean squared error from the general linear model.

The variance of individual predicted Y_{ijk} is estimated from

$$Var(Y_{ijk}) = [e^{(2Y_{ijk} + \sigma^2)}][e^{\sigma^2} - 1]. \quad (15)$$

Biggs and Funk (1988) found the bias introduced through use of s^2 for σ^2 to be minimal in the estimation of the mean egg count. A similar substitution is therefore made for σ^2 in Equation 15.

A re-examination of diver calibration data used for previous studies indicated that a simple linear model forced through the origin may be appropriate (personal communication, Fritz Funk, Alaska Department of Fish and Game, Juneau). After the 1994 field season, an ADF&G biometrician will re-evaluate the diver calibration model and recommend the most appropriate bias correction procedure.

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:

$$B' = \frac{\bar{W}S}{F(\bar{W}_f)} 10^3, \quad (16)$$

where

\bar{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,

$F(\bar{W}_f)$ = estimated fecundity at the average weight of females in the spawning population in an area, in numbers of eggs, and

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

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:

$$\begin{aligned}
\text{Var}(B') = & (10^3)^2 \left(\left[\frac{S}{F(\bar{W}_p)} \right]^2 \text{Var}(\bar{W}) \right. \\
& + \left[\frac{\bar{W}}{F(\bar{W}_p)} \right]^2 \text{Var}(S) \\
& + \left[\frac{\bar{W}S}{F(\bar{W}_p)^2} \right]^2 \text{Var}(F(\bar{W}_p)) \\
& + 2\text{Cov}(\bar{W}, S) \left[\frac{S}{F(\bar{W}_p)} \right] \left[\frac{\bar{W}}{F(\bar{W}_p)} \right] \\
& - 2\text{Cov}[\bar{W}, F(\bar{W}_p)] \left[\frac{S}{F(\bar{W}_p)} \right] \left[\frac{\bar{W}S}{F(\bar{W}_p)^2} \right] \\
& \left. - 2\text{Cov}[S, F(\bar{W}_p)] \left[\frac{\bar{W}}{F(\bar{W}_p)} \right] \left[\frac{\bar{W}S}{F(\bar{W}_p)^2} \right] \right)
\end{aligned}
\tag{17}$$

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 , $\text{Cov}(\bar{W}, S)$ and $\text{Cov}[S, F(\bar{W}_p)]$. Because the term involving $\text{Cov}[\bar{W}, F(\bar{W}_p)]$ has been shown to be very small in previous analyses and probably contributes little to $\text{Var}(B')$, these covariance terms will not be included in the estimate of $\text{Var}(B')$.

Herring Age, Weight, Length, Sex, and Fecundity:

This portion of the project is part of an existing agency program that is conducted annually by ADF&G. AWL information will be collected from major concentrations of herring spawning in each of the five spawn deposition summary areas (Baker et al. 1991a, Baker et al. 1991b, Wilcock et al. *in press*). Sampling generally occurs soon after concentrations of herring appear in nearshore areas and are accessible to purse seines. Samples will be taken periodically from most large 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 each commercial fishery and 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 in PWS (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. Therefore, we will collect fecundity samples in 1994 as a subsample of female herring from AWL samples and stratify by fish length. Egg and gonad weights will be measured and used to calculate average fecundity at the average female weight ($F(\bar{W}_f)$) from expression (16).

A fecundity sampling goal was set based upon the precision of the biomass estimate. Since the spawn deposition survey attempted to estimate spawning biomass with 95% confidence interval of no more than $\pm 25\%$ of the biomass estimate, we wanted the fecundity estimate to contribute no more than 1% to the confidence interval width. 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 in 1994 to maintain the coefficient of variation below 2.0%. In order to collect females over the range of 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 gonad weight will be assumed to be the equivalent of the weight of the ovaries removed from each female. Gonadal somatic index (GSI) will be defined as the percentage of total herring weight accounted for by gonad weight and will be calculated by dividing the gonad weight by body weight of each female sampled.

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

Sex ratio, S , will be calculated as the ratio of the number of herring of both sexes in AWL samples to the number of females. The binomial distribution is applicable to estimating the proportion, p , of females in AWL samples, where $S = 1/p$. The variance of S is

$$Var(S) = \frac{S^2(S-1)}{n}, \quad (18)$$

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

Fecundity for Biomass Estimates. Average fecundity for PWS will be estimated from a fecundity-weight relationship as $F(\bar{W}_f)$, and used in equation 16 to estimate biomass from spawn deposition. The variance of estimated average fecundities will be approximated by the variance of predicted means from the fecundity-weight linear regression (Draper and Smith 1981)

$$Var[F(\bar{W}_f)] = s^2 \left[\frac{1}{n} + \frac{1}{q} + \frac{(\bar{W}_f - \bar{WF})^2}{\sum (W_i - \bar{WF})^2} \right], \quad (19)$$

where

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

A linear 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.

Biggs et al. (*in press*) examined year and area differences in fecundity using analysis of covariance (ANCOVA) models that include all possible interaction terms. Area differences appeared relatively less important than year differences and average fecundity by area will not be estimated for 1994.

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 studies designed to test hypotheses about constraints to fishery production posed in the SEA plan. 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 most strongly correlated with sea surface temperatures 4 to 9 months prior to spawning. He also found that fecundity decreased as water temperatures increased.

Egg Loss Study

In the biomass estimation equation (1), T or total eggs is a direct multiplier. The total number of eggs estimated is corrected for egg loss or proportion of eggs removed prior to the spawn survey (equation 3, term R). Since the egg loss term directly affects the spawn deposition survey biomass estimate, it is important to improve the accuracy and definition of error associated with it. Prior to 1994, ADF&G has used a constant of 10% egg loss prior to surveys assuming that surveys are generally conducted 5-6 days after spawning as recommended in the literature (Haeghele et al. 1981, Blankenbeckler and Larson 1982).

New information reveals that egg loss rates are highly variable, site specific, and are generally higher than previously estimated. Jake Schweigert (personal communication, Canadian Department of Fisheries and Oceans, Nanaimo, B.C.) recently estimated average daily egg loss rates of 7.7 to 8.3% per day and an average total egg loss of 70% over the 14 day incubation period. Egg loss was studied during 1990 and 1991 in PWS (Biggs et al. *in press*) and an average daily egg loss rate of 2.1% and an average total egg loss of 50.4% over the 22.5 day incubation period were reported. These studies indicated that egg loss is a function of time and because it is variable between sites, a straight 10% correction factor irrespective of habitat type and number of days between spawning and surveys may not be appropriate. However, previous studies have not included collection of data to relate egg loss to habitat type, environmental conditions, or predation. The current study will include modifications to sampling design to improve understanding of these mechanisms behind egg loss. The revised egg loss information and model will be used to adjust spawn biomass estimates and improve the accuracy of stock assessment methods. Ultimately, this information and previous results can be used to build an embryo survival model as outlined in the SEA plan (project 94320).

Up to fifteen egg loss transects will be established in 1994 after herring spawn; 10 transects on Montague Island and 5 within the Northeast Area (Figure 1). These two areas were chosen to represent major spawning areas because herring consistently spawn every year in those areas. Habitat type within these major areas will be preliminarily defined prior to the field season and finalized when actual spawn distribution is known. Transects will be established perpendicular to shore following a compass course and will be randomly placed within each habitat type. Three sampling stations will be located along each transect line at three depths within the range of usual herring spawn (+1.65 m to -9.90 m; Figure 2). Based on previous egg loss and egg distribution information, sampling stations will be set at (1) 1.0 m above MLLW, (2) 1.0 m below MLLW, and (3) 3.0 m below MLLW. Station depths may be adjusted depending on actual egg deposition patterns in 1994. Depth will be determined using SCUBA diver depth gauges and later corrected for tide level. Each transect will be visited every three to four days.

A grid of 5 x 2 permanent 0.1 m² quadrats will be placed along the transect at each depth. Grids will be placed perpendicular to the transect and parallel to the shoreline. Permanent

grids will allow divers to estimate the number of eggs in the same grid over time. Divers will make estimates of egg density within each of the five 0.1 m² quadrats along the top row of the grid and the bottom row will serve as alternates in the case of the destruction of any in the top row.

At each visit and depth, divers will place a separate 0.1 m² quadrat off the end of the grid, estimate egg density within the quadrat, and collect all the eggs and vegetation within the quadrat for calibration samples. These diver calibration samples will be preserved and processed in the same manner as those collected for spawn deposition surveys.

An additional sample containing over 200 eggs, adjacent to the frames, will be haphazardly selected during each visit and depth. Within this small sample, live/dead ratios will be estimated and the eggs will be examined for any signs of egg desiccation and fungal infection and the incidence rate recorded. Just prior to hatch, a subsample of live embryos collected for live/dead examination will be immersed in preservative for later evaluation of morphological abnormalities and cytogenetics in the FY95 funding cycle. This information will be used to determine baseline levels of abnormalities according to habitat type, egg density, and egg distribution. This information is important both for distinguishing natural occurrences from oil spill effects and will be an important component of the survival model outlined in the SEA plan. Data on embryo survival and abnormalities collected in 1991 for NRDA Study 11 could be incorporated into the embryo survival model, but earlier data from 1989 and 1990 in PWS cannot be considered for baseline level definition due to oil spill effects.

The 1994 program will include the collection of detailed information on predation and wave loss. Within the 20 randomly placed transects predator exclusion frames will be placed at each of the three sampling depths. Three frames of approximately 1 m³ in volume will be placed at each depth: (1) a frame enclosed with small mesh that will retain all the eggs lost from wave action and at the same time exclude large visible predators, (2) a frame with mesh large enough to exclude avian predators, but allow for physical egg removal by waves, and (3) a control frame with no mesh. The total count of eggs within each frame will be estimated by each of three calibrated divers each time the site is visited (approximately every third or fourth day) and eggs and vegetation that have been physically dislodged will be gathered and recorded. In addition, live/dead ratios of remaining attached eggs will be recorded along with dates of hatch, any visible signs of egg desiccation, and any incidence of fungal infection. Half way through the incubation period or approximately 10 days after the mid-point date of spawn, three calibrated divers will estimate the eggs within approximately 1 m² patch of spawning area adjacent to and equivalent in area to that within the three frames and remove all the herring eggs, vegetation and all other animals. Eggs and vegetation will be preserved in Gilson's solution and all vertebrate and invertebrate animals will be frozen. Eggs, plants, and animals will be counted and recorded at the laboratory in Cordova. These data will contribute prey availability information directly to the study of avian predation on herring roe (Project 94320, part of SEA).

Physical measurements including air and water temperature, salinity, precipitation, wind speed and direction, and tide height will be collected at each site during each visit. Other measurements, such as gradient, substrate and vegetation will be collected once when the site is set up. Regional meteorological and oceanographic data will be obtained later from

shipboard surveys, moored instrumentation, and existing data products from government agencies (such as local measurements of wind shear). These measurements will be used to define habitat type and to model the effect of meteorological conditions on egg loss and embryo survival contingent upon funding in FY95.

Egg Loss Data Analysis. Systematic biases among divers and among vegetation types were found to exist in diver estimates from previous spawn deposition surveys in PWS (personal communication, Fritz Funk, Alaska Department of Fish and Game, Juneau). We assumed the same biases would exist in diver estimates for egg loss, and accounted for them as we had in spawn deposition surveys. Diver estimates of egg density will be approximately proportional to laboratory-enumerated counts. The model used to account for biases in diver estimates will be identical to the model described by equations (12) and (13).

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

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

where

- α = a constant,
- ADJ_{ijk} = adjusted egg density estimates,
- $trans_j$ = parameters representing the effect of transect j,
- $depth_k$ = parameters representing the effect of depth k,
- τ_{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 .

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

$$\log_e(ADJ_{ijk}) = \alpha + trans_j + depth_k + \tau_{jk}(days_{ijk}) + \epsilon_{ijk} \quad (23)$$

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)

$$\begin{aligned} \arcsin(s) = & \mu + \text{treat}_j + \text{depth}_k + \text{day}_i + \text{treat} * \text{depth}_{jk} + \\ & \text{day} * \text{treat}_{ij} + \text{day} * \text{depth}_{ik} + \text{day} * \text{treat} * \text{depth}_{ijk} + \\ & \text{trans}(\text{treat})_{ij} + \epsilon_{ijk} \end{aligned} \quad (24)$$

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 and predation. Additional analysis and modelling will be included in FY95 to determine the relationship of meteorological conditions and egg loss due to 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.

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Location

This project will be conducted entirely within PWS and it is expected that project results will directly affect the management of PWS herring fisheries (Figure 1). The communities directly affected that house fishermen, vessels involved in the fisheries, processing plants, and support services for the fisheries include Cordova, Seward, Valdez, and Whittier. The subsistence harvests of the native villages of Tatitlek and Chenega will also be directly

affected. Information gained in the egg loss portion of the project and associated predation study may be extremely valuable in the assessment of critical habitat and energy needs of migratory birds in PWS and throughout the rest of the state.

Technical Support

ADF&G regional and headquarters biometric staff will assist in project planning, review, and reporting for this project. They will also provide primary assistance for analysis of spawn deposition data and generation of biomass estimates using partial funding from this project. Additional biometric and modelling assistance for egg loss data analysis will be contracted through a Cooperative Service Agreement (CSA) or a Reciprocal Service Agreement (RSA).

Primary databases and analytical files will be stored on the local area network (LAN) of the Cordova ADF&G office and technical assistance for database management for FY94 will be provided by the Cordova office network administrator. Future database management needs will require additional funding for a research analyst or equivalent part time position beginning in FY95. In addition, we will coordinate with the SEA plan data managers to ensure that future incorporation and integration with their system can be accomplished.

Laboratory services will be completed at the Cordova ADF&G office with the exception of some collections of intertidal invertebrates and fishes, and samples collected for cytogenetic analysis. The intertidal predator samples will be transferred to the Copper River Delta Institute of the USFS to complete their research objectives. Analysis of cytogenetic samples will be subcontracted in FY95.

Contracts

Through a competitive bidding process, an 80 foot vessel will be chartered as a research platform for approximately 6 weeks to meet the data collection objectives of the project. This vessel will be shared by the avian predation project of the SEA plan (Project 94320).

Biometric and modelling assistance will be contracted for a period of 3 to 4 man-months through a sole-source CSA or RSA.

SCHEDULES

Spawn Deposition and Egg Loss:

Nov 93 - Feb 94	Initiate vessel charter bids and contract
	Secure divers, ensure certification requirements are met or in progress
	Complete data review and sample design for egg loss study

	Complete sample design for diver calibration
	Complete Detailed Project Description
Mar 94	Complete any necessary diver certifications
	Order laboratory supplies and field supplies
	Hire personnel to maintain and assemble dive gear
1-5 Apr 94	Complete all hiring of field personnel and arrange for arrival of divers
	Vessel arrives on site for inspection
1-4 Apr 94	HazMat, CPR/First Aid and Dive Safety training; Project orientation
	Set up laboratory
5-15 Apr 94	Initiate diving/field data collection (at onset of spawning)
	Set up egg loss sites and begin diving
1-15 May 94	Complete field activities
	Begin lab processing of calibration, fecundity, and egg loss samples
30 May 94	Complete data entry of diver estimates
May-Jun 94	Maintain, repair, and store gear
15 Jun 94	Complete calibration sample processing
30 Jun 94	Data entry of calibration samples
	Initiate data analysis
15 July 94	Complete egg loss sample processing and data entry
15 Aug 94	Preliminary biomass estimate
1 Sep 94	Finalize estimate of spawning biomass
15 Nov 94	Finalize projection of 1995 run biomass
Dec 94	Complete annual reports

EXISTING AGENCY PROGRAM

Existing programs within ADF&G that will contribute directly to this project include the ADF&G PWS Aerial Survey Program funded annually at approximately \$25K, the AWL Sampling Program funded in 1994 at \$23.9K, and the Cordova local area network with shared funding by all existing PWS programs. We will also be using 1.5 man-months of Fishery Biologist II and 1.5 man-months of Fish and Wildlife Technician II time from the management staff paid for by the Pound Fishery Monitoring Program funded at \$40K. Project planning and review will receive assistance from ADF&G biometric staff.

ENVIRONMENTAL COMPLIANCE/PERMIT/COORDINATION STATUS

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.

PERFORMANCE MONITORING

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) and NOAA's Auke Bay Laboratory reporting systems. 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.

The project leader and project staff is supervised by Steve Fried, Regional Research Coordinator, CFMDD, Anchorage Office. Dr. Fried has the ultimate authority and responsibility for this project.

COORDINATION OF INTEGRATED RESEARCH EFFORT

Project 94166 will be integrated closely with 94320, PWS SEA plan. One component of this plan, avian predation on herring spawn, will involve considerable sharing of resources. One technician will be housed aboard the spawn deposition contract vessel and share the research platform, skiffs, and man-power. Avian predation crews will collect synoptic information on bird abundance, behavior, and other data at our sampling transects. The two projects will share sample collection and laboratory processing duties for samples collected from these transects. The data management will be coordinated as outlined in the SEA plan for integration of results.

Project 94166 will also share information and resources with Project 94165, Herring Genetic Stock Identification in PWS. Samples for this project will be collected during AWL sampling as an agency contribution. Results from 94165 will be used to improve our knowledge of stock definition, improve recovery monitoring, and aid the formulation of fisheries harvest strategies.

Finally, integration of research will require data sharing and coordination with Project 94163, Forage Fish Influence on Injured Species. Because herring are an important forage species, their abundance and distribution information must be integrated with the composition, abundance, and distribution information of these other fish species. Because herring and other forage fish may be potential predators, competitors, and prey for each other at various stages throughout their life histories, understanding the population structure of these other species will improve our understanding of herring early life history and recruitment. In addition, herring and other forage fish stock dynamics information may lead to a better understanding of food availability, populations fluctuations, and breeding success of birds and mammals. Results from forage fish studies and spawn deposition surveys, as well as the

physical oceanographic components of the SEA plan will be essential for drafting a valid study design for herring larval advection studies beginning in FY95.

PUBLIC PROCESS

Following the dramatic herring population decline in 1993 coupled with the lack of ongoing research in 1993 and the continued documentation of oil spill injury, there has been a tremendous amount of public support for herring research from PWS communities and organizations. This support includes the Public Advisory Group (PAG) for the Trustee Council. The spawn deposition project has long been recognized by both commercial fishermen and fishery managers as an invaluable tool for stock assessment. In addition, the ecosystem approach to PWS studies adopted by the SEA planning group recognizes the commercial and ecosystem importance of herring and includes them as a co-target species for study along with pink salmon. This research planning effort was an intensive public review and planning process that occurred over a number of months and identified a number of key investigations to improve our understanding of the herring resource. The spawning biomass and egg survival results from spawn deposition surveys are necessary prerequisites to any additional ecosystem based studies of herring.

PERSONNEL QUALIFICATIONS

1. Project Leader - John Wilcock

John A. Wilcock, Herring Fisheries Research Biologist, Alaska Department of Fish & Game, P.O. Box 669, Cordova, Alaska 99574. **Education:** Bachelors of Science, Fisheries, University of Washington, 1978. **Professional Experience:** Fisheries Area Research Biologist, ADFG, 1992-1993; Fisheries Research Project Biologist, ADFG, 1982-1991; Fisheries Technician and Assistant Project Biologist, ADFG, Statewide Stock Biology Section, 1981-1982; Scientific Aide, Washington Department of Fisheries, 1979; Research Aide, Fisheries Research Institute, 1978-1979. **Research Projects:** EVOS injury to PWS herring, 1992-1993; Prince William Sound Eshamy District scale patterns analysis stock identification study, 1992-1993; Project Leader, Yukon River chum salmon scale pattern analysis feasibility study, 1988-87; Project Leader, Yukon River chinook salmon stock biology project 1982-1989. **Selected Publications:** Wilcock, J. A., *Annual Reports 1983-1987. Origins of Chinook Salmon in the Yukon River Fisheries*. Technical Fishery Reports. ADF&G, Juneau, Alaska; Wilcock, JA, TT Baker, ED Brown. *Stock Assessment and Management of Pacific Herring in Prince William Sound, Alaska, 1991*. Technical Fishery Report. ADF&G. Juneau, Alaska (1993). **Member:** American Fisheries Society, Alaska Chapter.

2. Project Co-Leader - Evelyn Brown

Evelyn D. (Biggs) Brown, 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 surveys for Prince William Sound, 1988, with historic overview.* ADFG Regional Informational Report, 2C88-07. Anchorage, Alaska. 45 p (1988). **Member:** American Fisheries Society, Alaska Chapter.

3. Biometric Support - David Evans, Biometrician I, CFMDD, ADF&G, Anchorage. 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 (Ontario, Canada), and a Master of Science in statistics from Oregon State University. David has worked with the Alaska Department of Fish and Game since October, 1991. His primary responsibility has been analysis of coded-wire-tag data from Prince William Sound. He has designed the statistical procedures and computer spread sheets used for inseason analysis of tag recovery data, has overseen most of the post season data analyses and has co-authored interim and final reports for the 1991 NRDA F/S Study #3, the 1992 Restoration Study 60C, and 1993 Restoration studies 93137 and 93184.

K. BUDGET

See attached

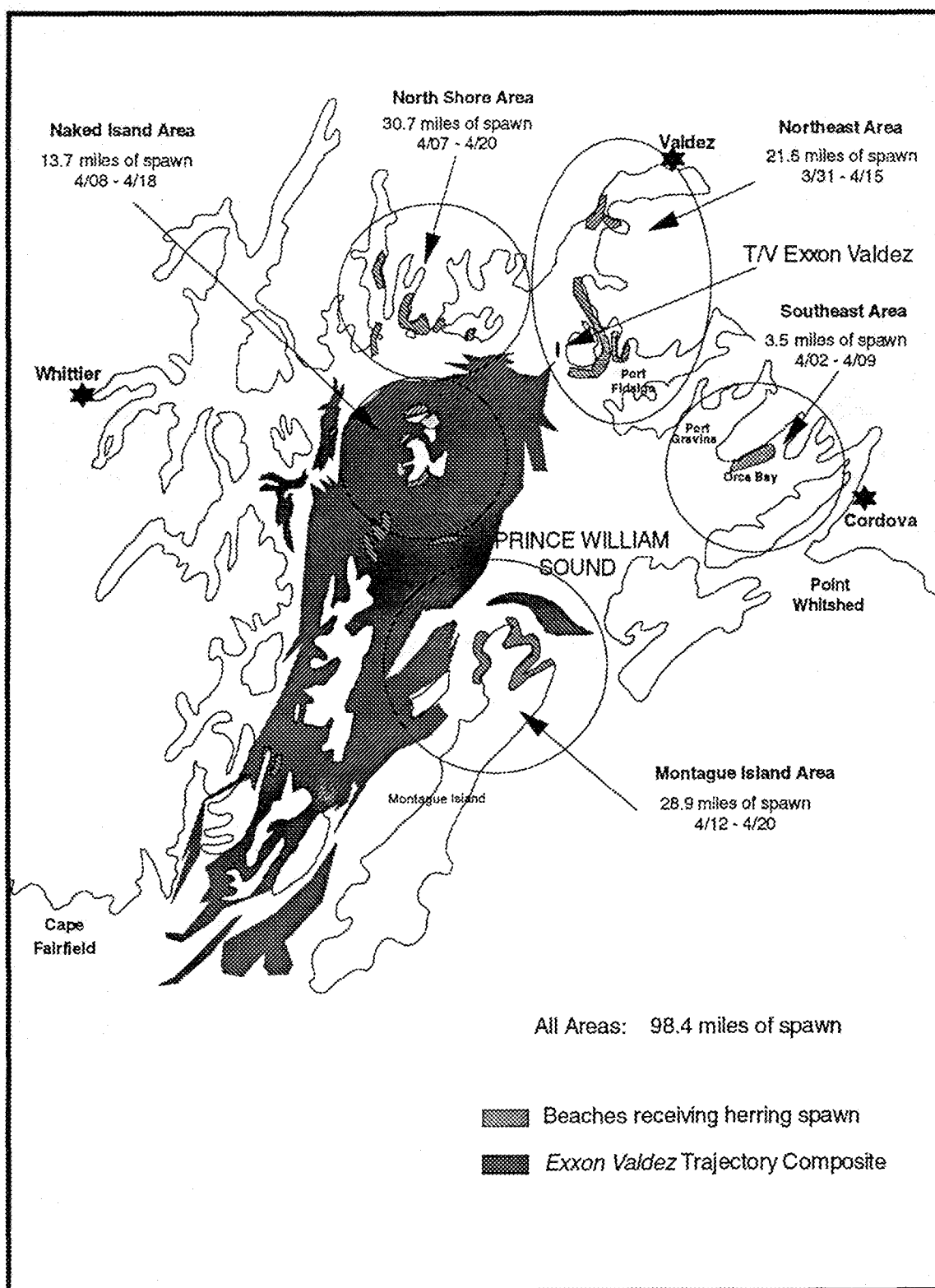
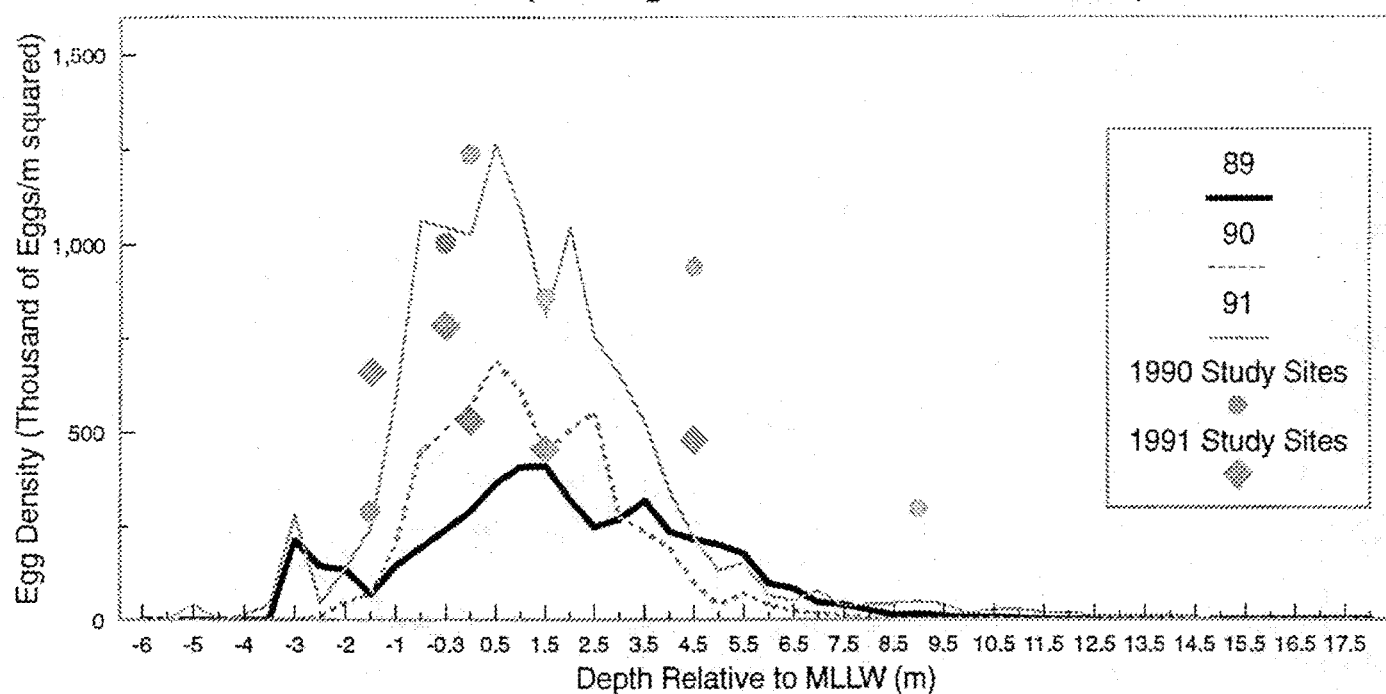


Figure 1. The Exxon Valdez oil spill trajectory composite and Prince William Sound herring spawn in 1989.

Oiled Areas (Montague Island and Naked Island)



Un-oiled Areas (Northeast and North Shore)

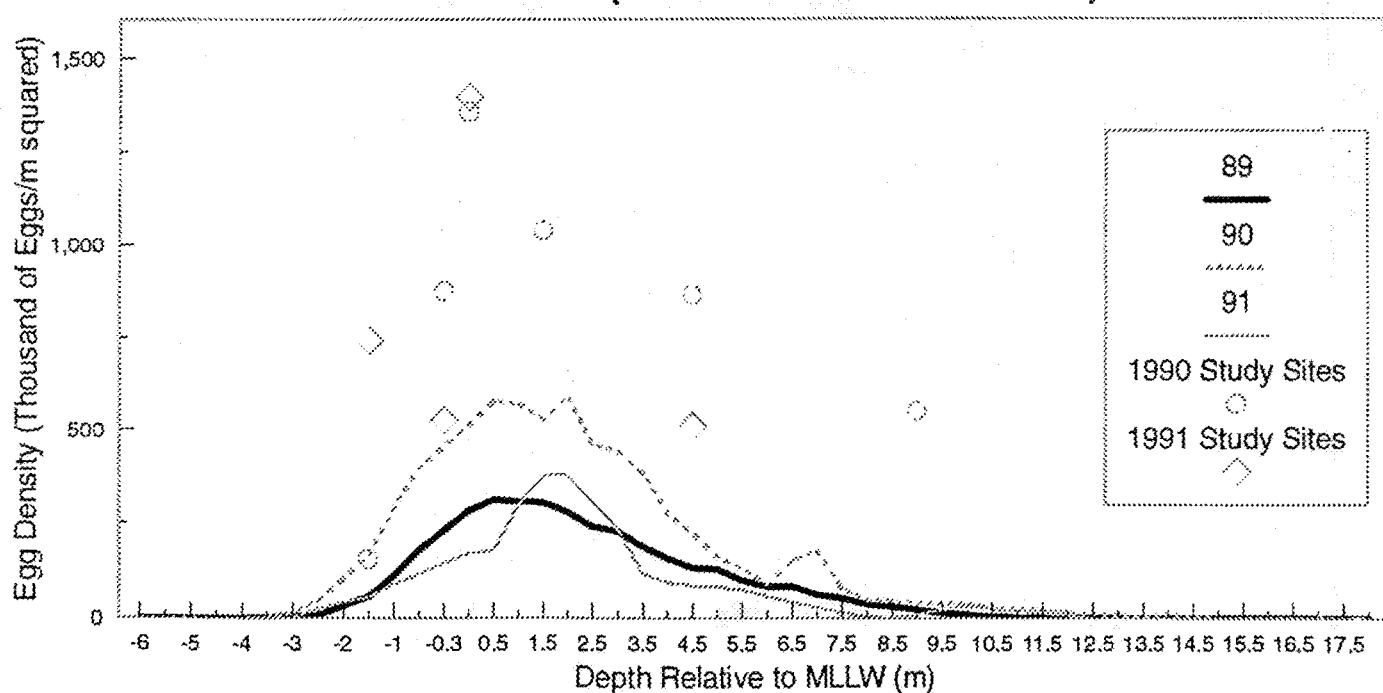


Figure 2. Distribution of herring egg density by depth and area in Prince William Sound during 1989, 1990, and 1991 (negative depths are intertidal).

EXXON VALDEZ OIL SPILL RECOVERY TRUST COUNCIL
 1994 Federal Fiscal Year Project Budget
 October 1, 1993 - September 30, 1994

Project Description: Herring Spawn Deposition & Reproductive Impairment - This project is designed to aid restoration of PWS herring resources through intensive management of commercial use. Scuba surveys are conducted to quantify herring spawn in areas of spawn identified through aerial surveys. Estimates of deposited spawn are combined with other biological information (age, sex, size, fecundity, etc.) to estimate the biomass of reproducing herring. Biomass estimates are used to forecast future returns and set harvest allocations.

Budget Category:	1993 Project No. Authorized FFY 93	'93 Report/ '94 Interim* FFY 94	Remaining Cost** FFY 94	Total FFY 94	FFY 95	Comment
Personnel	\$0.0	\$28.4	\$231.1	\$259.5	\$259.5	
Travel	\$0.0	\$0.8	\$7.1	\$7.9	\$7.8	
Contractual	\$0.0	\$16.0	\$98.9	\$114.9	\$117.8	
Commodities	\$0.0	\$12.4	\$20.7	\$33.1	\$23.6	
Equipment	\$0.0	\$0.0	\$3.9	\$3.9	\$3.9	
Capital Outlay	\$0.0	\$0.0	\$0.0	\$0.0	\$0.0	
Subtotal	\$0.0	\$57.6	\$361.7	\$419.3	\$412.6	
General Administration	\$0.0	\$5.4	\$41.6	\$47.0	\$47.2	
Project Total	\$0.0	\$63.0	\$403.3	\$466.3	\$459.8	
Full-time Equivalents (FTE)	0.0	0.5	4.0	4.5	4.3	
Dollar amounts are shown in thousands of dollars.						
Budget Year Proposed Personnel: Position Description		Reprt/Intrm Months	Reprt/Intrm Cost	Remaining Months	Remaining Cost	
See Individual 3A Forms for Personnel Details						
Personnel Total		0.0	\$0.0	0.0	\$0.0	
NEPA Cost: \$0.0						
*Oct 1, 1993 - Jan 31, 1994						
**Feb 1, 1994 - Sep 30, 1994						

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Project Number: 94166
 Project Title: Herring Spawn Deposition & Reproductive Impairment
 Lead Agency: AK Dept. of Fish & Game

FORM 2A
 PROJECT
 DETAIL

EXXON VALDEZ TRUSTEE COUNCIL
 1994 Federal Fiscal Year Project Budget
 October 1, 1993 - September 30, 1994

Project Description: Herring Spawn Deposition & Reproductive Impairment - This project is designed to aid restoration of PWS herring resources through intensive management of commercial use. Scuba surveys are conducted to quantify herring spawn in areas of spawn identified through aerial surveys. Estimates of deposited spawn are combined with other biological information (age, sex, size, fecundity, etc.) to estimate the biomass of reproducing herring. Biomass estimates are used to forecast future returns and set harvest allocations.

Budget Category:	1993 Project No. Authorized FFY 93	'93 Report/ '94 Interim* FFY 94	Remaining Cost** FFY 94	Total FFY 94	FFY 95	Comment
Personnel	\$0.0	\$23.4	\$143.5	\$166.9	\$166.9	Total Egg loss \$30.9
Travel	\$0.0	\$0.8	\$4.0	\$4.8	\$4.8	\$0.0
Contractual	\$0.0	\$6.0	\$58.9	\$64.9	\$67.8	\$17.5
Commodities	\$0.0	\$3.0	\$6.3	\$9.3	\$9.2	\$2.0
Equipment	\$0.0	\$0.0	\$3.9	\$3.9	\$3.9	\$0.0
Capital Outlay	\$0.0	\$0.0	\$0.0	\$0.0	\$0.0	\$0.0
Subtotal	\$0.0	\$33.2	\$216.6	\$249.8	\$252.6	\$50.4
General Administration	\$0.0	\$3.9	\$25.6	\$29.6	\$29.8	\$5.9
Project Total	\$0.0	\$37.1	\$242.2	\$279.4	\$282.4	\$56.3
Full-time Equivalents (FTE)	0.0	0.4	2.3	2.7	2.7	
Dollar amounts are shown in thousands of dollars.						
Budget Year Proposed Personnel:		Reprt/Intrm Months	Reprt/Intrm Cost	Remaining Months	Remaining Cost	FFY 95 costs may vary depending upon FFY 94 results. No report cost are requested as this is a new project for '94. NEPA Cost: \$0.0 *Oct 1, 1993 - Jan 31, 1994 **Feb 1, 1994 - Sep 30, 1994
Position Description						
Intrm 1 Fisheries Biologist III		0.5	\$3.0	4.0	\$27.5	
3 Fisheries Biologist II (Intrm- 1 FB II)		2.0	\$10.6	6.5	\$40.8	
5 Fisheries Technician II (Intrm- 2 FWT)		0.5	\$1.6	8.3	\$32.3	
2 Biometrician I (Intrm- 1 Biom I)		1.0	\$4.8	5.0	\$23.8	
1 Program Manager		0.5	\$3.4	1.0	\$6.7	
1 Fisheries Technician III		0.0	\$0.0	2.8	\$12.4	
Personnel Total		4.5	\$23.4	27.6	\$143.5	

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Project Number: 94166
 Project Title: Herring Spawn Deposition & Reproductive Impairment
 Sub-Project: Field Surveys
 Agency: AK Dept. of Fish & Game

FORM 3A
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EXXON VALDEZ STEE COUNCIL
 1994 Federal Fiscal Year Project Budget
 October 1, 1993 - September 30, 1994

Travel:		Reprt/Intrm	Remaining
Intrm	Spawn deposition:		
	1 RT Juneau/Cordova (air fare \$650 + \$150 per diem)	\$0.8	\$0.0
	3 RT Homer/Cordova (air fare \$500 + 3.5 days per diem @ \$150/day)	\$0.0	\$2.0
	4 RT Anchorage/Cordova (air fare \$350 + 4 days per diem @ \$150/day)	\$0.0	\$2.0
Egg loss:	None		
Travel Total		\$0.8	\$4.0
Contractual:			
Intrm	Spawn deposition:		
	Diver physicals (5 @ \$425)	\$2.1	\$0.0
	Equipment maintenance/repair	\$1.0	\$0.0
	Dive master class for 3 dive leaders for annual recertification	\$1.2	\$0.0
	Vessel charter (6 divers/25 days)	\$0.0	\$37.5
	Skill fuel/maintenance	\$0.0	\$2.5
	Dive equipment repair/maintenance	\$0.0	\$1.8
	Hazmat training	\$0.0	\$0.9
	Shipping and postage	\$0.0	\$0.2
Egg loss:	Basic diving	\$0.3	\$0.0
	Dive annual recertification (3 @ \$200)	\$0.6	\$0.0
	Diver physicals (2 @ \$425)	\$0.8	\$0.0
	Vessel charter (15 days X \$1.0/day)	\$0.0	\$15.0
	Fuel	\$0.0	\$1.0
Contractual Total		\$6.0	\$58.9

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Project Number: 94166
 Project Title: Herring Spawn Deposition & Reproductive Impairment
 Sub-Project: Field Surveys
 Agency: AK Dept. of Fish & Game

FORM 3B
SUB-
PROJECT
DETAIL

EXXON VALDEZ TRUSTEE COUNCIL
 1994 Federal Fiscal Year Project Budget
 October 1, 1993 - September 30, 1994

Commodities:		Reprt/Intrm	Remaining
Intrm Spawn deposition:			
Upgrade Cordova lab to meet OSHA minimum standards for storage and disposal of hazardous materials		\$3.0	\$0.0
Network operation/maintenance		\$0.0	\$0.5
Software upgrades		\$0.0	\$0.2
Office/lab supplies (pencils, paper, formalin, etc.)		\$0.0	\$1.0
Food for field camps for 20 days x 4 people x \$20/day		\$0.0	\$1.6
Skiff repair materials		\$0.0	\$1.0
Egg loss:			
Food for field camps for 15 days x 5 people x \$20/day		\$0.0	\$1.5
Field sampling supplies		\$0.0	\$0.5
Commodities Total		\$3.0	\$6.3
Equipment:			
Spawn deposition:			
Chemical storage locker for hazardous materials		\$0.0	\$1.5
Dive gear replacement (2 dry suits)		\$0.0	\$2.4
Egg loss:			
None			
Equipment Total		\$0.0	\$3.9

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Project Number: 94166
 Project Title: Herring Spawn Deposition & Reproductive Impairment
 Sub-Project: Field Surveys
 Agency: AK Dept. of Fish & Game

FORM 3B
 SUB-
 PROJECT
 DETAIL

EXXON VALDEZ STEE COUNCIL
 1994 Federal Fiscal Year Project Budget
 October 1, 1993 - September 30, 1994

Project Description: Herring Spawn Deposition & Reproductive Impairment - This project is designed to estimate long term damage from oiling by predicting genetic damage to germ tissue in laboratory experiments. Herring will be exposed to oil and assessed for somatic genetic damage and other biological damage and compared to chemical uptake experiments to infer genetic damage to germ tissue.

Budget Category:	1993 Project No. Authorized FFY 93	'93 Report/ '94 Interim* FFY 94	Remaining Cost** FFY 94	Total FFY 94	FFY 95	Comment
Personnel	\$0.0	\$5.0	\$87.6	\$92.6	\$92.6	
Travel	\$0.0	\$0.0	\$3.1	\$3.1	\$3.0	
Contractual	\$0.0	\$10.0	\$40.0	\$50.0	\$50.0	
Commodities	\$0.0	\$9.4	\$14.4	\$23.8	\$14.4	
Equipment	\$0.0	\$0.0	\$0.0	\$0.0	\$0.0	
Capital Outlay	\$0.0	\$0.0	\$0.0	\$0.0	\$0.0	
Subtotal	\$0.0	\$24.4	\$145.1	\$169.5	\$160.0	
General Administration	\$0.0	\$1.5	\$15.9	\$17.4	\$17.4	
Project Total	\$0.0	\$25.9	\$161.0	\$186.9	\$177.4	
Full-time Equivalents (FTE)	0.0	0.1	1.7	1.8	1.6	
Dollar amounts are shown in thousands of dollars.						
Budget Year Proposed Personnel: Position Description	Reprt/Intrm Months	Reprt/Intrm Cost	Remaining Months	Remaining Cost		
Interm Principal Investigator GS 12	0.9	\$5.0	4.0	\$23.4		
Chemist GS 9	0.0	\$0.0	8.0	\$33.9		
Fish Biologist GS 7	0.0	\$0.0	8.0	\$22.8		
Program Manager GS 12	0.0	\$0.0	0.9	\$7.5		
Personnel Total	0.9	\$5.0	20.9	\$87.6		
					NEPA Cost:	\$0.0
					*Oct 1, 1993 - Jan 31, 1994	
					**Feb 1, 1994 - Sep 30, 1994	

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Project Number: 94166
 Project Title: Herring Spawn Deposition & Reproductive Impairment
 Sub-Project: Laboratory Verification
 Agency: National Oceanic & Atmospheric Admin.

FORM 3A
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EXXON VALDEZ TRUSTEE COUNCIL
 1994 Federal Fiscal Year Project Budget
 October 1, 1993 - September 30, 1994

Travel:	Reprt/Intrm	Remaining
2 RT Anchorage/Juneau for meetings (air fare \$450 + 1.5 days per diem @ \$225/day)	\$0.0	\$1.6
One report presentation at a scientific meeting (yet to be determined)	\$0.0	\$1.0
1 RT to PWS associated with ADF&G field activities	\$0.0	\$0.5
Travel Total	\$0.0	\$3.1
Contractual:		
Intrm Purse seine contract to collect adult herring for lab exposure to hydrocarbons to evaluate possible larval abnormalities and g	\$10.0	\$0.0
Genetic analyses contract to look for possible genetic injury to larval herring (cost based on previous contracts)	\$0.0	\$17.0
Mixed-function oxidase histological and chemical analyses to determine exposure to hydrocarbons (cost based on previous contracts)	\$0.0	\$17.0
Analyses of bile for presence of metabolites of petroleum hydrocarbons (cost based on previous sample analysis costs)	\$0.0	\$6.0
Contractual Total	\$10.0	\$40.0

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Project Number: 94166
 Project Title: Herring Spawn Deposition & Reproductive Impairment
 Sub-Project: Laboratory Verification
 Agency: National Oceanic & Atmospheric Admin.

FORM 3B
 SUB-
 PROJECT
 DETAIL

EXXON VALDEZ LITIGATION COUNCIL
 1994 Federal Fiscal Year Project Budget
 October 1, 1993 - September 30, 1994

Commodities:		Reprt/Intrm	Remaining
Intrm	Wet lab supplies	\$2.5	\$1.0
	Chemistry lab supplies	\$4.4	\$3.0
	Solvents for GC/MS	\$2.5	\$0.0
	Culturing supplies (media, glassware, tubing)	\$0.0	\$7.4
	Culturing setup (lights, temperature recording and monitoring)	\$0.0	\$3.0
Commodities Total		\$9.4	\$14.4
Equipment:			
Equipment Total		\$0.0	\$0.0

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Project Number: 94166
 Project Title: Herring Spawn Deposition & Reproductive Impairment
 Sub-Project: Laboratory Verification
 Agency: National Oceanic & Atmospheric Admin.

FORM 3B
 SUB-
 PROJECT
 DETAIL

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EXXON VALDEZ TRUSTEE COUNCIL
FY 94 DETAILED PROJECT DESCRIPTION

A. COVER PAGE

Project title: Pigeon Guillemot Recovery Monitoring

Project ID number: 94173

Project type: Research/Monitoring

Name of project leader(s): Dennis Marks (acting)

Lead agency: U.S. Fish and Wildlife Service

Cooperating agencies: NOAA, ADFG, SEA study cooperators

Cost of project/FY 94: \$200K

Cost of project/FY 95: \$350K, plus cost of FY94 write-up, \$55.2K

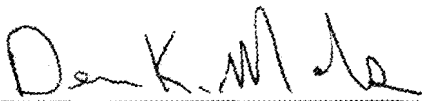
Cost of Project/FY 96 and beyond: \$290K per year

Project Start-up/Completion Dates: 2/94, 4 years

Geographic area of project: Prince William Sound

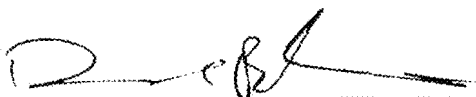
Name of lead agency project manager: David Irons

Project Leader:


Dennis K. Marks (Acting), Wildlife Biologist
DOI-FWS (Migratory Bird Management)

Date: 3/18/94

Project Manager:


David B. Irons, Wildlife Biologist
DOI-FWS (Migratory Bird Management)

Date: 3/18/94

B. INTRODUCTION

The Prince William Sound (PWS) pigeon guillemot (*Cephus columba*) population has been in decline since the 1970s. Guillemots were heavily impacted by the *Exxon Valdez* oil spill; an estimated 2,000 to 3,000 were killed throughout the spill zone (Piatt et al. 1990). This figure includes 33% of the 1991 estimated Prince William Sound (PWS) population. Data from waterbird surveys indicated that the PWS summer guillemot population declined from about 15,000 in 1972 to 6,600 in 1991 and 4000 in 1993 (Islieb and Kessel 1973; Klosiewski and Laing 1992; Agler et al. 1993). A survey was conducted in 1993 to locate pigeon guillemot colonies in PWS (Sanger and Cody MS). Approximately 1,800 guillemots were recorded at 160 colonies throughout PWS and a total of 3,077 guillemots were recorded in nearshore waters during the colony surveys. Based on colony catalog data, the dispersion throughout PWS appears to have remained about the same since the early 1970's, though the population has decreased dramatically (USFWS). This spatial pattern supports data demonstrating an ongoing decline in the breeding population, rather than the idea that there has been movement of guillemots between areas within PWS. Latent effects from the spill may yet be impacting the population since guillemots do not breed until they are three or four years old.

Relationships between forage fish abundance and seabird colony size and reproductive success have long been observed, and severe declines in food fish have been blamed for declines and catastrophic failures of seabird colonies (Monaghan et al. 1989). Low reproductive rates in seabirds have been attributed to limitations in the ability to deliver food to young (Lack 1968, Ricklefs 1983). Long foraging trips by seabirds have been correlated with low chick feeding rates and subsequent breeding failures (Irons 1992, Hamer in press) and nesting attempts, clutch size and hatching and fledging success are all directly affected by food availability (Murphy et al. 1991). Activity budgets and colony attendance of breeding adults, chick growth, breeding (hatching and fledging) success and adult survivorship may all be affected by prey abundance and availability. Extreme weather and other factors can influence reproduction parameters, but chick growth (and maximum weight before fledging), activity budgets and colony attendance appear to be sensitive measures of prey availability and foraging success (Cairns 1987).

Pigeon guillemots are well suited as samplers of the nearshore community as they have one of the most diverse diets found among seabirds, including midwater (e.g., Pacific sand lance, *Ammodytes hexapterus* and Pacific herring, *Clupea harengus pallasii*), and bottom fish (e.g., blennies and sculpins) and invertebrates. Individual birds appear to specialize on a much narrower range of prey and have been observed to maintain unique diet preferences despite the use of obvious and abundant different prey by neighbors (Drent 1965, Kuletz 1983). Guillemot reproductive success has been shown to reflect abundance of prey; Ainley and Boekelheide (1990) found that when juvenile rockfish were unavailable during the chick rearing period, the fledging weights of pigeon guillemots were low regardless of hatching date, and that mean

fledgling weight was in fact highly correlated to the proportion of rockfish in the diet.

Egg and chick predation is a potential factor influencing guillemot reproductive success in PWS. Of 61 pigeon guillemot nests investigated in Queen Charlotte Islands, B.C. (Vermeer et al. 1993) 71.2% of egg loss and 58.6% of chick loss was due to predation, and was the greatest cause of egg and chick loss. Most of the known predation was avian. In PWS common predators on pigeon guillemot eggs, chicks and adults at the nest probably include common ravens (*Corvus corax*), northwestern crows (*Corvus caurinus*), black-billed magpies (*Pica pica*), glaucous-winged gulls (*Larus glaucescens*), mink (*Mustela vison*) and river otters (*Lutra canadensis*). The primary difference in reproduction that was observed following the spill on Naked Island was a decreased nesting success rate which was due to an increased rate of nest loss from predation, during the chick stage.

In the Naked Island area, the number of guillemots along oiled shorelines was less than expected following the spill even though few breeding birds were in the area when the oil came through (Oakley and Kuletz MS). The most likely mechanism may be related to the guillemot's use of beach rocks at their colony sites. Guillemots rest and socialize on intertidal rocks at their colony sites and may have come in contact with oil at that time. In turn, eggs could have died after oil was transferred from parent's breast feathers to eggs. Due to intertidal habits, guillemots may still be exposed to traces of oil remaining in intertidal areas.

To determine if pigeon guillemot productivity is limited by food, predation or oiling, we will initiate a multi-year study at several colonies in PWS. In the first year, diet, foraging behavior, predation rates, egg oiling and reproductive success will be compared among colonies in different areas of PWS. Relative forage fish abundance will be compared to reproductive parameters. This study will help identify reasons for the guillemot population decline in PWS.

Information on the foraging ecology of the pigeon guillemot will contribute to the development of PWS ecosystem trophic models. If food is limiting, guillemot recovery will depend on the health of populations of sand lance, capelin, herring, pollock, rockfish, sculpin, blennies and a variety of other bottom fish. The relative accessibility of the guillemot for reproduction and food studies make them a useful organism for understanding needs of other piscivorous marine birds (e.g., marbled murrelets, puffins, arctic terns, kittiwakes) and mammals (e.g., harbor seals).

C. PROJECT DESCRIPTION

1. Resources and/or Associated Services:

The injured resource targeted by this project is the pigeon guillemot. By providing information on guillemot diet, foraging behavior and reproductive success in conjunction with the forage fish study (94163) and the Sound Ecosystem Assessment

[SEA project, 94320], this study will provide information on whether forage fish availability limits guillemot reproduction. Additionally, the diet information will aid in defining the trophic pathways needed to develop an ecosystem model for the PWS marine environment. This project will benefit guillemots by identifying factors which may be causing the population to continue to decline in PWS. Knowledge gained by monitoring guillemot populations may apply to other seabirds in PWS, especially marbled murrelets (*Brachyramphus marmoratus*), parakeet auklets (*Cyclorhynchus psittacula*), tufted puffins (*Fratercula cirrhata*) and horned puffins (*Fratercula corniculata*). These bird species have nests which are much less accessible and are therefore probably much more difficult to study than the guillemot.

2. Relation to Other Damage Assessment/Restoration Work:

Exxon Valdez oil spill damage assessment studies conducted in 1989-1990 (Bird Study 9) focused on guillemot prey and productivity. In 1991 and 1992 work was conducted ancillary to the R15 restoration project on marbled murrelet nesting habitat. Restoration Study 93034, completed in 1993, surveyed the entire shoreline of PWS and mapped guillemot colonies and estimated numbers of breeding and non-breeding birds.

Similar goals of determining forage fish availability and feeding behavior are of interest to the Murrelet Prey and Foraging Habitat Study (94102), and will allow us to coordinate programs and personnel to improve upon study design and allow for greater coverage and more efficient data collection. Both of these studies will also be closely linked to data collection and analysis conducted by SEA (94320) study (especially the juvenile herring and forage fish components) and the forage fish (94163) study. The forage fish study will collect fish and bird abundance data at two study areas used by the murrelet prey and foraging and pigeon guillemot recovery monitoring projects. The forage fish studies will provide data on fish species, abundance, and distribution (both spatially and temporally) early in the season, and provide temperature, bathymetric, and other oceanographic measurements that affect fish distribution and availability.

3. Objectives:

To determine if reproductive success of PWS pigeon guillemots is limited by food, predation and/or persistent oil contamination we will:

- a. Determine nesting chronology, chick growth rates, fledging weight and reproductive success at Naked and Fool Island and determine if differences between areas exist.
- b. Determine chick diet, chick provisioning rate and foraging bout duration information at several colonies to determine variability in PWS, differences between areas and correlations with prey distribution.

- c. Document foraging areas and determine abundance of forage fish at foraging sites (in conjunction with forage fish study). Determine relative abundance of benthic fish and invertebrates.
- d. Identify predators and predation rates of eggs, chicks and incubating adults and identify nest types most vulnerable to predation.
- e. Determine if there is persistent oiling of guillemot eggs.

4. Methods:

a. Study Area

We will study guillemots nesting at several sites. Naked Island, where extensive work has been done in the past, and Fool Island, where nests were identified in 1993, will be the focus of 1994 work on reproductive success. Additional sites with large guillemot colonies, including Jackpot Island, Bligh Island, Harriman Fjord, Blackstone Bay and Passage Canal (Figure 1) will be prepared for 1994 chick feeding observations and reproductive success work in 1995 by identifying nest sites, banding birds, and preparing blinds for observations. Guillemots nest in talus crevices, cliff crevices and cliff-edge burrows which will be located during early incubation and chick rearing periods.

b. Data Collection

Daily weather measurements will be taken at each area and will include maximum and minimum air temperatures, wind speed and temperature. Sea conditions will be estimated and rated on a scale of 1-5.

Chick growth and reproductive success (objective a) - Reproductive success will primarily be monitored at Naked and Fool Island. Methods developed and refined in past years on Naked Island will be used (Oakley and Kuletz MS). Thirty to 50 nests will be monitored at Naked and Fool Islands. Nest checks will be conducted every 4-5 days to determine number of eggs laid, approximate hatching date and hatching success, chick growth, and maximum and fledging weight. Nest checks will involve checking for incubating adults, banding and measuring chicks. Wing chord, tarsi and culmen length will be measured to the nearest 1 mm and chick weight (chicks will be placed in measuring bags and weighed with Pesola hand-held scales) to the nearest 1 g. Fledging weight is the weight measured within one week of fledging.

Chick diets, provisioning rates and foraging bout duration (objective b) - At Naked and Fool Islands chick provisioning watches will be conducted every 2-3 days for approximately 6 daylight hours. At other sites watches will be performed opportunistically. Guillemots will be observed with spotting scopes and binoculars from blinds assembled near colonies. Fish will be identified to species, where

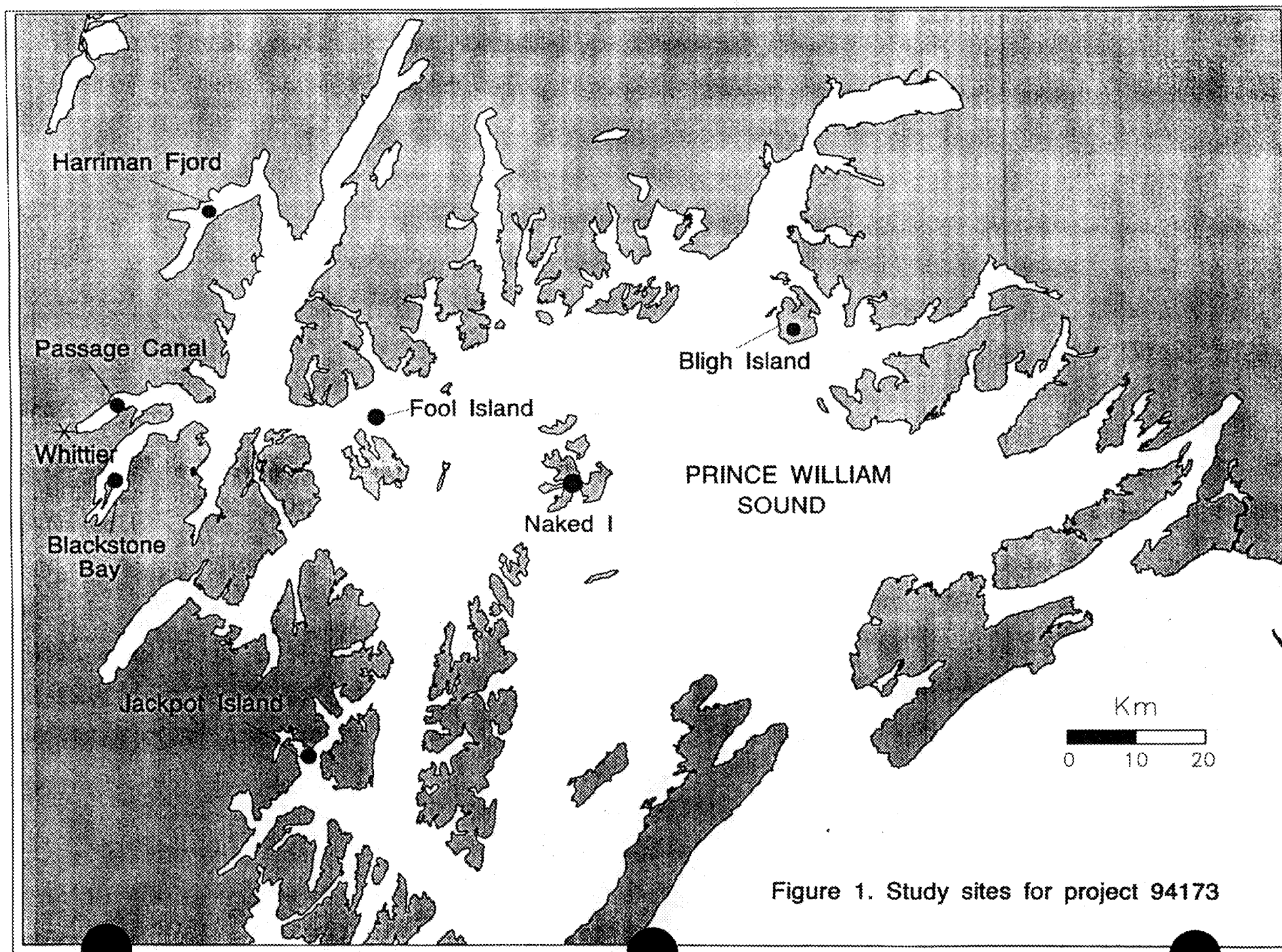


Figure 1. Study sites for project 94173

possible, and will at least be classified as either a schooling or a bottom fish. Eleven prey categories based on taxonomy and the ability to distinguish certain groups of prey species will be used here when species identification is not possible (Oakley and Kuletz MS). Prey size will be judged by comparing prey organism to the bill of the guillemot, and will be recorded to the nearest 1/2 bill length. Additional information on diet will be obtained opportunistically, for example, fish identified from nests and otoliths taken from fecal samples collected from nests. Ancillary diet data from the SEA study and NOAA forage fish studies will be collected opportunistically from on-board observers; a concentrated bout of observations will be made on foraging grounds near colony sites when hydroacoustically outfitted vessels are surveying in the immediate area of these sites.

Foraging bout durations will be recorded while observing individual birds from blinds. Forage bout duration is defined as the time from when a guillemot leaves the nest to when it returns again. We will examine distribution of foraging areas in relation to colony sites. Guillemots will be visually followed to foraging areas where information on prey type, capture rate, dive times and forage bout length at foraging grounds will be collected to evaluate foraging grounds. Using compasses and landmarks and maintaining radio contact between the observer in the blind and a person in an inflatable boat, flight directions of incoming and outgoing birds will be recorded and birds will be tracked to forage areas. On foraging areas we will measure physical factors (sea surface temperature, sea surface salinity, depth) to relate food abundance and guillemot distribution to oceanographic characteristics.

Blood lipid analysis has shown differences between diets of marine mammals (Iverson 1993). In order to gather more information on guillemot diets without sacrificing birds, we will investigate developing this technique for determining marine bird diets. We will remove a small amount of blood from the wing tissue of adults captured for banding and from juvenile, pre-fledgling, guillemots.

Fish abundance (objective c) - To characterize guillemot foraging habitat in an effort to relate habitat and prey abundance with chick provisioning we will sample benthic prey and schooling forage fish. Minnow traps have been successful at capturing bottom fish and invertebrates in previous years and will be deployed here at depths of between 3-25 m for 8-24 h at a time (Oakley and Kuletz MS). They will be set randomly in areas of guillemot foraging. The forage fish study (94163) will sample the water column adjacent to guillemot colonies using hydroacoustics to determine relative prey abundance and trawls to determine species composition. The guillemot study may also use small otter trawls to obtain an estimate of prey abundance; transects will be conducted in the areas in, and adjacent to foraging guillemots. Throw nets may be used opportunistically to identify species in the water column not identified by other means.

Predation estimates (objective d) - Live traps will be set on occasion to identify predators. Traps will be set in vegetation above the colonies as well as on rocks within the boundaries of the colony. Other sign, including animal tracks and scat, will

be identified. Rates of predation will be estimated primarily from nest observations. Predation on eggs and chicks is determined by examination of egg or chick remains. When eggs or chicks disappear and it is obvious that they could not have fallen out of sight, predation is assumed. Predator identification on egg or animal remains is sometimes possible; mink and other carnivores have characteristic chew marks; corvid holes can sometimes be identified in egg shell fragments.

Oiling (objective e) - Sealed sample jars for use for hydrocarbon analysis will be taken into the field. Any unhatched eggs will be collected, using hydrocarbon sample collection protocol, and sent to Texas A & M laboratories for analysis.

c. Data Analysis

Hatching success will be defined as the average number of chicks hatched or fledged per nest for all nests found during the egg stage; fledging success is the mean number of chicks fledged per nest for all nests whether found in the chick or egg stage (Oakley and Kuletz MS). Chick growth rates, provisioning rates and foraging trip duration will be compared between areas using t-tests and compared among three or more colonies using one-way Analysis of Variance. Hatching success, fledging success, reproductive success and predation rates will be compared among colonies using chi-square tests.

5. Location:

The Naked Island group supports approximately 27% of the PWS guillemot population, and is located on a shallow shelf about 24 km from the mainland. Naked Island will be included among the study sites because of the high density of guillemots, the past data available for comparison, and the detailed knowledge of the colonies. The other sites were chosen based on 1) the existence of a large number of accessible nest sites, and 2) an attempt to study a variety of locations around PWS, with various habitat types and bathymetric characteristics. All precautionary measures will be taken so that no habitat will be adversely affected by this project and every effort will be made to minimize the impact on individual animals and communities. Boats and field materials will be transported through Whittier, Alaska, 55 mi southeast of Anchorage on Passage Canal, PWS. Use of docks and other facilities and proximity to study sites make this an excellent staging area for this project.

6. Technical Support:

Texas A&M University will provide technical support by analyzing unhatched eggs for hydrocarbon.

7. Contracts:

We will contract, by competitive bid, a barge to transport camp equipment and supplies, including fuel, to the Naked Island study site. This camp will be coordinated with the marbled murrelet study (94102) to use the same barge deliveries. A multi-year contract has been awarded to Texas A & M for hydrocarbon analysis.

Also in conjunction with the marbled murrelet study, we will contract with the University of Alaska, Fairbanks, Institute of Marine Science (IMS) for identification of fish species not able to be identified by USFWS personnel. The blood lipid analysis will also be contracted.

D. SCHEDULES

Apr 1-May 10, 1994: Hire biological technicians, conduct safety training for all field personnel, prepare boats and other field gear, prepare final surveys protocols

May 10-20, 1994: Finalize preparations for field, transport boats, other field equipment and field personnel to Whittier, AK. Construct blinds at colony sites, track birds to foraging grounds (primarily by visually following them from nest areas to foraging grounds) and make observations (fish species, dive times, etc.). Locate nests at colonies, band birds.

May 20-Aug 10, 1994: Conduct nest burrow checks for egg production, egg survival, hatching dates and chick survival and growth. Chick feeding observations (from blinds) identifying fish species and approximate size, recording feeding rate. We will continue to make observations on foraging grounds.

Aug 11-25: Continue to make observations and measurements of birds on nests.

Aug 26-Sep 31, 1994: Remove blinds, transport boats and gear back to Anchorage. Clean and maintain field equipment. Data entry and preliminary analysis.

Oct 1-Jan 15: Continue data analysis and writing.

Jan 15: Draft report to FWS Oil Spill Coordinator.

Feb 1, 1995: Draft report due to Chief Scientist.

Apr 30, 1995: Final report complete.

E. EXISTING AGENCY PROGRAM

Currently, the FWS has no ongoing programs to monitor the PWS pigeon guillemot population. The only ongoing work in PWS using base funding is on kittiwakes.

F. ENVIRONMENTAL COMPLIANCE/PERMIT/COORDINATION STATUS

Based on a review of the CEQ regulation 1500-1508, this study has been determined to be categorically exempt from the requirements of NEPA in accordance with 40 CFR 1508.4.

G. PERFORMANCE MONITORING

The project manager, D. Irons, will oversee the project and assign responsibilities to other agency personnel if other key staff are unable to complete the project. In the event that the one of the (associate) project leaders leaves before the project's completion, the other leader will assume responsibility for completion. The project manager will be available for additional assistance, if needed.

Quality assurances will be provided in the field by hiring experienced personnel. In addition to the principal investigator, at least two people with knowledge of pigeon guillemots and PWS will be returning.

Personnel will be trained to identify fish at a distance with actual samples, after identification training with plates and photos has been completed. One of the principal investigators will be present with each of the field groups. All data will be entered into a computer database and archived at USFWS. Reports will be submitted to USFWS Oil Spill Coordinator for internal review, followed by the Trustee Council peer review.

H. COORDINATION OF INTEGRATED RESEARCH EFFORT

This project is integrally related to the NOAA forage fish project (94163). The two projects will collect complementary data that will be shared between the two projects, to determine if food is limiting recovery of pigeon guillemots. The pigeon guillemot restoration monitoring study and the murrelet food and foraging habitat study (94102) are also coordinated so that field camps, transport, communication and observations can be shared and the forage fish study will provide data for both the pigeon guillemot and murrelet studies. Through cooperation with the SEA study, bird observers will be aboard research vessels and will collect both bird data to complement fish distribution data. Close coordination with the forage fish and the murrelet study at least will continue throughout the life of this study because all three are sharing data integral to all studies.

I. PUBLIC PROCESS

This project has been reviewed by the Exxon Valdez Oil Spill (EVOS) Public Advisory Group, the EVOS Trustee Council and has been published for public review. The short proposal for this project was published as part of the proposed FY94 work plan

and made available for public comment. The final study proposal (this document) will be available for public and peer review.

J. PERSONNEL QUALIFICATIONS

Migratory Bird Management Project Manager: David Irons received his Ph. D. from the U. of CA, Irvine in 1992. His dissertation was on the foraging ecology and breeding biology of the black-legged kittiwake. The field work for this study was conducted in Prince William Sound. Irons received his M. S. from Oregon State University in 1982 where he studied foraging behavior of glaucous-winged gulls in relation to the presence of sea otters. Irons conducted marine bird and sea otter surveys in PWS in 1984 and 1985. He has been studying kittiwakes in PWS for 11 years and completed the EVOS kittiwake damage assessment study. Irons has overseen several seabird studies in the past few years including a marine bird and sea otter survey in PWS and in Cook Inlet, a seabird monitoring study on Little Diomed Island, and a cost of reproduction study on kittiwakes. Irons has authored and co-authored several reports and publications on seabirds and has made several presentations at scientific conferences on seabirds.

Pertinent reports and publications:

Irons, D.B. Submitted to Auk. Size and productivity of black-legged kittiwake colonies in Prince William Sound, Alaska before and after the T/V *Exxon Valdez* oil spill.

Irons, D.B. In preparation. Foraging site fidelity and tidal rhythms in individual Black-legged Kittiwakes.

Irons, D.B. In preparation. Flexible foraging behavior in seabirds: short-term buffer and long-term tradeoff?

Irons, D.B. In preparation. The role of food availability in sibling aggression and brood reduction of the Black-legged Kittiwake.

Irons, D.B. 1992. Factors Affecting Black-legged Kittiwake reproductive success. Unpublished Ph.D. Dissertation.

Irons, D.B., R.G. Anthony, and J.A. Estes. 1986. Foraging strategies of Glaucous-winged Gulls in a rocky intertidal community. *Ecology* 67:1460-1474.

Hatch, S.A., G.V. Bryd, D.B. Irons, and G.L. Hunt. 1993. Status and ecology of kittiwakes in the North Pacific Ocean. Pages 140-153 in editors, K. Vermeer, K.T. Briggs, K.H. Morgan, D. Siegel-Causey, The status, ecology, and conservation of marine birds of the North Pacific. Can. Wildl. Serv. Spec. Publ., Ottawa, Canada.

Hogan, M.E. and D.B. Irons. 1986. Waterbirds and marine mammals. in M.J. Hameedi and D.G. Shaw, editors. Environmental management of Port Valdez, Alaska: scientific basis and practical results. Springer-Verlag, New York.

Vermeer, K., and D.B. Irons. 1991. The Glaucous-winged Gull on the Pacific Coast of North America. *Acta Twentieth Congressus Internationalis Ornithologici*:2378-2383.

Acting Principal Investigator: Dennis Marks received a B.S. from the U. of CA, Irvine in 1979 and completed his M.S. at the University of Oregon Institute of Marine Biology where he studied the feeding ecology of several species of bottom fish. In 1990 he participated in the marbled murrelet and pigeon guillemot damage assessment studies. In 1991 he was an integral part of the marbled murrelet restoration study. In 1992 and 1993, he supervised two marbled murrelet restoration studies and co-authored several reports and publications on the results of these studies. Prior to these studies, Mr. Marks spent several years coordinating field projects on the west coast and in S.America.

Pertinent reports and publications:

Kuletz, K.J., D.K. Marks, and N.L. Naslund. MS. At-sea abundance and distribution of marbled murrelets in the Naked Island Area in summer, 1991 and 199 Annual Progress Report. U.S. Fish and Wildlife Service, Anchorage, Alaska. 32pp.

Kuletz, K.J., N.L. Naslund and D.K. Marks. MS. Identification of marbled murrelet nesting habitat in the *Exxon Valdez* oil spill zone: Restoration Project R15 Annual Progress Report. U.S. Fish and Wildlife Service, Anchorage, Alaska. Four Chapters with 7 appendices. 150 pp.

Marks, D.K., K.J. Kuletz and N.L. Naslund. In review. Marbled murrelet surveys in Prince William Sound, Alaska: Surveying for marbled murrelet nesting habitat in remote areas. Proceedings of the Pacific Seabird Group's Marbled Murrelet Symposium. 27pp.

Marks, D.K. 1987. Effects of protracted disturbance on the food of the English Sole (*Parophrys vetulus* Knerr). M.Sc. Thesis. University of Oregon, 1987. 78pp.

Associate Principal Investigator: Mary Cody graduated from the University of Michigan in 1987 and has been with the FWS Division of Migratory Bird Management since 1989. Following a season working on the effect of the spill on black oystercatchers, she joined the 1990 USFWS marbled murrelet and pigeon guillemot damage assessment studies. In 1991, she conducted extensive habitat and murrelet surveys for the marbled murrelet restoration study in PWS and continued pigeon guillemot work on Naked Island. In 1992, Ms. Cody supervised pigeon guillemot and marbled murrelet research on Afognak Island, Alaska and assisted on seabird surveys there. In 1993 she was associate principle investigator for the PWS pigeon guillemot colony survey, and has co-authored several publications and reports on pigeon guillemots, marbled murrelets and other seabirds.

Pertinent Reports and Publications:

Andres, B.A., and M.B. Cody. MS. Effects of the *Exxon Valdez* oil spill on black oystercatchers breeding in Prince William Sound, Alaska. Bird Study 1 Final Report. U.S.Fish and Wildlife Service, Anchorage.

Cody, M.B., and G.A. Sanger. 1994. Survey of pigeon guillemot colonies in Prince William

- Cody, M.B., and T.P. Gerlach. 1993. Distribution and activity levels of marbled murrelets at coastal and inland sites on Afognak Island, Alaska. Biological Report. U.S. Fish and Wildlife Service, Anchorage, Alaska. 15pp.
- Cody, M.B., J. Fadely, and T.P. Gerlach. 1993. Population estimates of nesting seabirds along the northern and southwestern coast of Afognak Island, Alaska. Biological Report. U.S. Fish and Wildlife Service, Anchorage, Alaska. 33pp.
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- Fadely, J., T.P. Gerlach, and M.B. Cody. 1993. Distribution and population estimates of marine birds in the near-shore and pelagic waters of Afognak Island, Alaska. Biological Report. U.S. Fish and Wildlife Service, Anchorage, Alaska. 18pp.
- Kuletz, K.J., D.K. Marks, N.L. Naslund and M.B. Cody. In review. Marbled murrelet activity in four forest types at Naked Island, Prince William Sound, Alaska. Proceedings of the Pacific Seabirds Group's Marbled Murrelet Symposium. Northwestern Naturalist.
- Kuletz, K.J., D.K. Marks, N.L. Naslund, N.G. Stevens and M.B. Cody. MS. Information Needs For Habitat Protection: Marbled Murrelet Habitat Identification. U.S. Fish and Wildlife Final Report. 110pp.
- Naslund, N.L., K.J. Kuletz, M.B. Cody, and D.K. Marks. In review. Tree and habitat characteristics and behavior at fourteen marbled murrelet tree nests in Alaska. In Proceedings of the Pacific Seabird Group's marbled murrelet symposium, 1993. Northwestern Naturalist.
- Sanger G.A. and M.B. Cody. MS. Survey of pigeon guillemot colonies in Prince William Sound, Alaska. U.S. Fish and Wildlife Service Final Report. 58pp.
- Sharp, W., and M.B. Cody. MS. Effects of the Exxon Valdez oil spill on black oystercatchers in Prince William Sound, Alaska.

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- Irons, D.B. 199 Aspects of foraging behavior and reproductive biology of the black-legged kittiwake. Ph.D. diss., Univ. of California, Irvine, Irvine, Calif. 143 pp.
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- Klosiewski, S. P. and K. K. Laing. MS. Marine bird populations of Prince William Sound, Alaska, before and after the *Exxon Valdez* oil spill. NRDA Bird Study No. U.S. Fish and Wildlife Service, Anchorage, Alaska.
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- 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. *Journal of Animal Ecology* (1991), 60, 515-534.
- Oakley, K.L. and K.J. Kuletz. MS. Population, reproduction and foraging ecology of pigeon guillemots at Naked Island, Prince William Sound, Alaska, before and after the *Exxon Valdez* oil spill. Bird study No. 9. U.S. Fish and Wildlife Service, Anchorage, Alaska.
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- Vermeer, K. 1980. The importance of timing and type of prey to reproductive success of rhinoceros auklets (*Cerorhinca monocerata*) *Ibis*.
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EXXON VALDEZ TRUSTEE COUNCIL
 1994 Federal Fiscal Year Project Budget
 October 1, 1993 - September 30, 1994

Project Description: This project is designed to monitor pigeon guillemot population recovery in Prince William Sound. Breeding populations will be censused at all major colonies and compared with 1993 data. Productivity, diets, and predation will be determined and compared at the four largest colonies along oiled and unoiled shorelines, including oiled Naked Island, the only place in Prince William Sound where guillemots were previously studied before and after the Exxon Valdez Oil Spill.

Budget Category:	1993 Project No. Authorized FFY 93	'93 Report/ '94 Interim* FFY 94	Remaining Cost** FFY 94	Total FFY 94	FFY 95	Comment
Personnel	\$0.0	\$0.0	118.6 \$122.0	118.6 \$122.0	47.0 \$120.0	FFY 95 costs are to write the report for the field work conducted in FFY 94.
Travel	\$0.0	\$0.0	\$5.7	\$5.7	\$2.0	
Contractual	\$0.0	\$0.0	32.5 \$20.6	32.5 \$20.6	\$0.0	
Commodities	\$0.0	\$0.0	\$13.3	\$13.3	\$0.0	
Equipment	\$0.0	\$0.0	9.5 \$9.0	9.5 \$9.0	\$0.0	
Capital Outlay	\$0.0	\$0.0	\$0.0	\$0.0	\$0.0	
Subtotal	\$0.0	\$0.0	\$179.6	\$179.6	47.0 \$120.0	
General Administration	\$0.0	\$0.0	\$21.5	\$21.5	6.2 \$20.0	
Project Total	\$0.0	\$0.0	\$201.1	\$201.1	55.2 \$140.0	
Full-time Equivalents (FTE)	0.0	0.0	2.9	2.9	0.2	
Dollar amounts are shown in thousands of dollars.						
Budget Year Proposed Personnel:		Reprt/Intrm Months	Reprt/Intrm Cost	Remaining Months	Remaining Cost	
Position Description						
Principal Investigator GS-11 Biologist		0.0	\$0.0	7.0	\$41.0	
Wildlife Biologist GS-6 Biotech		0.0	\$0.0	6.0	\$19.0	
Supervisory Biologist PROJECT MANAGER		0.0	\$0.0	2.0 3.0	9.0 \$8.0	
4 Biological Techs GS-5		0.0	\$0.0	16.0 32.0	32.0 \$55.2	
Expeditor		0.0	\$0.0	2.0 1.0	5.6 \$2.0	
Program Manager		0.0	\$0.0	2.0 0.2	\$12.0	
Personnel Total		0.0	\$0.0	34.7	118.6 \$122.0	
					NEPA Cost:	\$0.0
					*Oct 1, 1993 - Jan 31, 1994	
					**Feb 1, 1994 - Sep 30, 1994	

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Project Number: 94173
 Project Title: Pigeon Guillemot Recovery Monitoring
 Agency: Dept. of Interior, Fish & Wildlife Service

FORM 2A
 PROJECT
 DETAIL

EXXON VALDEZ TRUSTEE COUNCIL
 1994 Federal Fiscal Year Project Budget
 October 1, 1993 - September 30, 1994

Travel:	Reprt/Intrm	Remaining
Portage to Whittier - vehicle and boat trailer (2 trips @ \$714/trip)	\$0.0	\$1.4
Portage to Whittier (3 trips - 6 people @ \$16/trip)	\$0.0	\$0.3
Field per diem (6 people x 110 days x \$3/day)	\$0.0	\$2.0
Anchorage to Cordova/Valdez (4 trips - air fare \$210/trip + 2 days per diem @ \$150/day)	\$0.0	\$2.0
Travel Total	\$0.0	\$5.7
Contractual:		
Hydrocarbon analysis of 10 unhatched eggs	\$0.0	\$8.0
Aircraft charter for supplies (4 trips x 2 camps x 150 lbs/camp x \$0.40/lb) + (4 trips x 2 camps x \$50 landing fee/trip)	\$0.0	\$0.8
Aircraft charter to survey outer coast of Hinchinbrook and Montague Islands (2 charters x 7 hours x \$125/hour + \$450/charter)	\$0.0	\$157
Freight for fuel delivery to remote locations in Prince William Sound	\$0.0	\$5.0
Mandatory safety, first aid, CPR, and wilderness survival training - ⁷⁵⁰ 6 people x \$100 /person)	\$0.0	4.5 \$100
Boat repair	\$0.0	\$5.0
Warehouse rental - 1 month	\$0.0	9.0 \$50
LIPID ANALYSIS		6.0
Contractual Total	\$0.0	32.5 \$29.6

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Project Number: 94173

Project Title: Pigeon Guillemot Recovery Monitoring

Agency: Dept. of Interior Fish & Wildlife Service

FORM 2B

PROJECT
DETAIL

EXXON VALDEZ TRUSTEE COUNCIL
 1994 Federal Fiscal Year Project Budget
 October 1, 1993 - September 30, 1994

Commodities:		Reprt/Intrm	Remaining
Food (6 people x 110 days x \$10/day)		\$0.0	\$6.6
Boat fuel (2 boats x 20 miles/day x 110 days x 1 mpg x \$1.40/gallon)		\$0.0	\$6.2
Camp supplies (tarps, stoves, stools to replace existing worn-out equipment)		\$0.0	\$0.5
Commodities Total		\$0.0	\$13.3
Equipment:			
Miscellaneous boat equipment		\$0.0	\$3.0
Other miscellaneous logistical equipment		\$0.0	\$2.0
			6.5
Equipment Total		\$0.0	9.5

07/14/93

1994

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Project Number: 94173
 Project Title: Pigeon Guillemot Recovery Monitoring
 Agency: Dept. of Interior, Fish & Wildlife Service

FORM 2B
PROJECT
DETAIL

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94185

**NO DETAILED PROJECT DESCRIPTION REQUIRED --
PROJECT IS A CONTINUATION OF
PREVIOUSLY APPROVED WORK**

BRIEF PROJECT DESCRIPTION FOLLOWS

EXXON VALDEZ OIL SPILL PROJECT DESCRIPTION

Title: Coded-wire Tagging of Wild Pink Salmon for Stock Identification in Prince William Sound

Project Number: 94185

Lead Agency: ADF&G

Cooperating Agency: None

Cost of Project, FY94: \$286.0K

Cost of Project, FY95: \$286K

Project Startup Date: October 1993

Duration: 2 years

Geographic Area: Prince William Sound

INTRODUCTION

Wild stocks of pink salmon (*Oncorhynchus gorbuscha*) are a critical component of the entire Prince William Sound (PWS) ecosystem. Extensive seaward migrations of pink salmon fry serve as both the dominant predators on zooplankton populations and as important prey for other fishes and birds. Historically, an average of 10 to 15 million pink salmon return from the high seas to spawn in PWS's hundreds of streams. These migrations are vital in sustaining a wealth of species that depend upon pink salmon to transport nutrients and energy from feeding grounds in the north Pacific to nearshore waters, freshwater streams, and upland ecosystems of PWS. In the past three years, the number of adult pink salmon returning to PWS have dramatically declined. Total returns of pink salmon have fallen from 1991's record high return of over 40 million fish to a near-record low return in 1992. Preliminary 1993 results indicate that this year's pink salmon return will continue this downward trend despite the strong parent year return in 1991 and continuous, steady, hatchery production from the four PWS pink salmon hatcheries.

The Alaska Department of Fish and Game's (ADF&G) intent to restore future PWS pink salmon escapements to historic levels will require both conservative and precise fisheries management strategies. Restoration efforts based upon inseason evaluation and manipulation of the commercial fishing fleet depends upon the manager's ability to identify and selectively reduce harvests on injured stocks. Management actions designed to protect wild pink salmon from overharvest in fishing districts with weak returns is the best option presently available to restore oil-impacted stocks from the southwest district of PWS. Stock in that district have experienced higher egg mortalities, larval deformities, and lower juvenile growth rates than stocks from unoiled streams and hatcheries. There is also evidence that oiled stocks may also have persistent genetic injury which has resulted in reduced egg survival in generations following the spill. Natural recovery from oil-related injuries will be hindered by extensive exploitation by the commercial fishing fleet targeting on hatchery

stocks which in recent years have dominated the total return. The dominance by hatchery fish of the total return has further compounded the complexity of making precise management decisions that would positively benefit individual wild stocks. Evidence from Natural Resource Damage Assessment (NRDA) Fish/Shellfish (F/S) Study #3 in 1991 and 1992 suggests that spawning populations of wild fish in PWS were subjected to a high degree of straying by hatchery populations. Extensive straying by tagged wild stocks was also documented both years. The magnitude and range of straying by both hatchery and wild pink salmon stocks may be the primary influence in the success or failure of restoration efforts directed at wild stocks.

This project will provide marked wild fish of known origin for recovery in the commercial catch, natal and non-natal spawning streams, and hatchery broodstock collections. Stock specific harvest information from coded-wire tag recoveries will provide inseason estimates of stock abundance, timing and eventually total return and survival for tagged stocks. Stock specific information will be used to establish optimal exploitation rates, investigate straying, and evaluate current and future restoration efforts.

Alternative approaches considered included applying tags to wild fish at more locations while foregoing total outmigration enumeration. This approach was rejected. Without total enumeration of a stream's outmigration, tag recoveries would not be representative of the entire population.

PROJECT DESCRIPTION

Previous NRDA studies (F/S #1, #3) demonstrated the technical feasibility of applying coded-wire tags to wild pink salmon fry. Recoveries of tagged fish from the commercial catch and from numerous streams have demonstrated that accurate estimates of stock abundance, timing, survival, and straying can be obtained using this technology. Accurately estimating these parameters will be necessary in order to gauge the success of management actions designed to restore injured populations.

A. Resources and/or Associated Services

The potential biological and economic benefits that would be attained from consistently strong returns of wild pink salmon provide compelling arguments for improving our understanding of the resource. The ability of managers to maintain healthy stocks of wild pink salmon will directly benefit the resource and indirectly other species that rely on the injured resource. Strong wild stock returns to all districts of PWS would enable the commercial fishing fleet to fully utilize the hatchery enhanced component of the pink salmon return and any wild stock surplus, without putting injured stocks at further risk. Strong returns would also benefit the diversity of species that fully utilize pink salmon.

Project Description

B. Objectives

The primary objective of the wild stock tagging project is to provide marked fish of known origin for eventual recovery in either the commercial catch or the escapement. Specific objectives are to:

1. Enumerate and characterize the outmigration of wild pink salmon fry from two streams during both an even and an odd year.
2. Tag a representative subsample of pink salmon fry at each location, each season.

C. Methods

The total outmigration of wild pink salmon fry from two streams will be enumerated using fyweirs. A representative subsample will be marked and tagged with half-length coded-wire tags. A minimum of 100,000 fry per stream will be tagged.

D. Location

Two representative spawning streams, one in eastern PWS and one in the southwest district will be selected as study streams. Selection will depend upon a stream's 1993 escapement and our ability to totally enumerate the outmigration at that location.

E. Technical Support

Minimal technical support will be needed to complete the desired objectives of this project. Tag code validation and data archiving by the ADF&G Tag Lab will be the primary technical support needed for this project.

F. Contracts

Site selection may involve streams surrounded by private landowners or federally protected lands. Lease agreements may need to be established depending upon site selection.

SCHEDULES

October 1993 - February 1994: Study site selection and preparation, equipment preparation, seasonal personnel recruitment.

March 1994: Field work begins. Camps and fry weirs set up.

April 1994 - June 1994: Coded-wire tagging representative subsamples of wild pink salmon fry from two streams for later recovery from the commercial catch and the escapement (Projects 94-184 & 94-192).

FY 95: Same timeline and objectives as FY94.

Overall project supervision will be under the direction of the PWS Area Research Biologist. Hiring, training, and coordinating all field activities will be under the direction of a Fisheries Biologist. Field activities will be performed by seasonal Fish and Wildlife Technicians. The field work for this project will take place in the spring, both in 1994 and 1995. Winter conditions dominate the weather in PWS during this time of year. Field camp startup in March will require vessel support as will camp breakdown at the end of the project. Weekly supply flights are planned to support activities at the two field camps.

EXISTING AGENCY PROGRAM

This project will not receive any other agency contributions or support during FY 1994. The wild stock coded-wire tagging project is a stand-alone project. Results from the 1994 tagging program will be seen in the 1995 adult pink salmon return.

ENVIRONMENTAL COMPLIANCE/PERMIT/COORDINATION STATUS

The ADF&G will obtain Title 16 permits for both intertidal fry weirs. All sampling of salmon fry is covered by an ADF&G biological collection permit. Necessary lease agreements with private land owners will be arranged by project personnel and reviewed for approval by the ADF&G Administration's Division of Leasing and the Department of Law. Proposed camps and weirs are not permanent structures and will be removed at the termination of the project.

The National Oceanic and Atmospheric Administration (NOAA) is lead Federal agency for National Environmental Act (NEPA) compliance for this project. This project meets NOAA agency requirements for Categorical Exclusion from the NEPA process.

Project Description

PERFORMANCE MONITORING

Personnel policy, purchasing practices, field camp operations, safety procedures and project administration will be in accordance with the ADF&G Division of Commercial Fisheries Management and Development Division Standard Operating Procedures Manual. Data collection and quality control procedures will be similar to those used for the wild stock tagging component of NRDA F/S Study #3 and in accordance with ADF&G Tag Lab database requirements. These procedures have undergone thorough review by ADF&G biometrics staff and by the Restoration Team peer review process. A report of the success of management actions based on this data will be prepared.

FY94 BUDGET (\$K)

	ADF&G
Personnel	176.8
Travel	1.5
Contractual	38.0
Commodities	35.0
Equipment	5.5
Capital Outlay	<u>0.0</u>
Subtotal	256.8
General Administration	<u>29.2</u>
Project Total	286.0
NEPA Compliance	0.0

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EXXON VALDEZ TRUSTEE COUNCIL
FY 94 DETAILED PROJECT DESCRIPTION

Project title: Oil Related Egg and Alevin Mortalities

Project ID number: 94191

Project type: Monitoring and Research

Name of project leaders: Samuel Sharr, Alaska Dept. Fish and Game
Jim Seeb, Alaska Dept. Fish and Game
Jeep Rice, National Marine Fisheries Service

Lead agency: Alaska Department of Fish and Game

Cooperating agencies: National Marine Fisheries Service
Other Cooperating Parties: Washington State University

Cost of project/FY 94: \$873.5
Cost of project/FY 95: \$741.2
Cost of Project/FY 96 and beyond: \$359.5

Project Start-up/Completion Dates: October 1, 1994 to September 30, 1996

Geographic area of project: Prince William Sound

Project leaders:

<u>Sam Sharr</u> Sam Sharr (ADF&G) By Brian Bue	<u>3/2/94</u> Date
<u>Jim Seeb</u> Jim Seeb (ADF&G) By Brian Bue	<u>3/2/94</u> Date
<u>Jeep Rice</u> Jeep Rice (NMFS) By Brian Bue	<u>3/2/94</u> Date

Agency project managers:

<u>Joseph R. Sullivan</u> Joe Sullivan (ADF&G)	<u>3/3/94</u> Date
<u>Bruce Wright</u> Bruce Wright (NMFS) By Brian Bue	<u>3/3/94</u> Date

B. INTRODUCTION

Summary

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 have 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 and functional sterilization of affected individuals, reducing the 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.

An increase in physiological dysfunction above that which would normally occur clearly would result in a reduction in production potential. A persistent decline of this nature could render present restoration efforts inadequate, as historical levels of spawning escapement would be insufficient to sustain a harvestable wild pink salmon population.

The purpose of this study is to continue to monitor the recovery of pink salmon embryos and fry in the field, provide laboratory verification of the field results presented by Sharr et al. (1994a and 1994b) and Bue et al. (in press), and test the hypothesis that exposure of pink salmon to a polluted incubation habitat will result in the functional sterilization of these animals at sexual maturity. In this study we will (1) survey the same streams examined during the Natural Resource Damage Assessment (NRDA) process for pink salmon eggs and preemergent fry in order to monitor recovery, (2) collect pink salmon gametes from oiled and non-oiled streams in western Prince William Sound (PWS) and incubate them under controlled conditions to evaluate the effect of physical stream characteristics upon the damages observed in the field, (3) utilize controlled laboratory exposures to fertilized eggs in a simulated inter-tidal gravel environment in order to mimic actual environmental exposures (link NRDA Study FS2), and (4) examine embryos and fry from both the field and laboratory work for presence of genetic aberrations.

History

Pink salmon eggs and fry incubating in the oiled intertidal spawning areas in PWS in 1989, 1990, 1991, and 1992 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 gravels 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 pink salmon egg mortalities in the intertidal zones of oiled streams while no difference between oiled and non-oiled streams was detected above mean high tide. Elevated egg mortalities in oiled streams were again detected in the 1990 brood year, but only in the highest intertidal spawning zone (Sharr et al. 1994a, and 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 egg 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 egg mortalities in oiled streams during the fall of 1991 survey. Furthermore, significant differences in egg mortality occurred at all tidal zones, including the area above mean high tide. Clearly, the elevated egg 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 egg mortalities in 1991 may be the result of genetic damage acquired during development after fertilization in 1989. Elevated egg mortalities at all tidal zones in oiled streams were again detected during the fall of 1992 survey (Sharr et al. 1994b and 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 sampling design (Sharr et al. in prep).

The hypothesis of genetic damage is consistent with previous laboratory experiments on the effects of crude oil on early life stages of fish and with other NRDA field observations. 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 eggs and larvae exposed to very low aqueous doses (1 ul oil/l seawater) of oil. Moles et al. (1987) confirmed that pink salmon eggs 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 reproductive fitness of a significant proportion of exposed individuals.

Genetic damage induced by genotoxins can be classified into two general categories: damage to the DNA molecule itself caused by nucleotide base substitutions, deletions, or additions (microlesions); and changes in chromosome number or structure (macrolesions). Chemical agents that induce mutations in DNA are also likely to produce cytologically recognizable chromosome damage expressed as structural changes or "aberrations" (Evans

1976). Flow cytometry is a cytogenetic technique that detects the visible effects of DNA macrolesions and will be the primary method used in this study for detecting genetic damage.

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). 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). This method allows for rapid and sensitive processing of large numbers of cells per individual and for timely analysis of many samples. The ability to quantify the cellular characteristics for many individuals in a short period of time greatly reduces lab costs over traditional cytogenetic analyses while providing greater statistical power for hypothesis testing.

Information gained from this study will provide resource managers insight to the magnitude and persistence of damages sustained by wild pink salmon due to EVOS. Efforts to restore damaged pink salmon populations depend upon the fishery manager's abilities to identify sources of reduced survival and to monitor their persistence. Information on the potential of long term oil exposures to cause genetic damage is needed so spawning escapement goals can be reevaluated and adjusted if necessary. In addition, verification of the genetic hypothesis would provide the first evidence that reproductive capacity of fish exposed to chronic or acute sources of oil pollution would be compromised.

C. PROJECT DESCRIPTION

This project is composed of three parts: (A) a recovery monitoring component which will continue to collect field information on pink salmon eggs and preemergent fry in order to observe recovery in the natural systems, (B) a laboratory fertilization component that will expose fertilized eggs from oiled and unoled streams in Prince William Sound to identical incubation environments, and (C) a laboratory oil exposure component that will expose fertilized eggs to an incubation environment contaminated with crude oil, rear surviving fry to maturity, and check their gametes for viability. Components B and C work together to verify the 1989 through 1993 field findings of Sharr et al. (1991, 1994a, 1994b, and in prep) and Bue et al. (in press). Differences in survival between groups in component B will be unrelated to incubation environment indicating problems with gamete quality. Differences in gamete viability between groups in component C will be related to oil exposure, and demonstrate the probable cause for effects observed in component B.

1. Resources and/or Associated Services:

This study will investigate pink salmon in Prince William Sound, Alaska, and pink salmon from Lover's Cove Creek in southeastern Alaska. Results of this study will have major implications with respect to the natural ecosystem and the economy of the PWS area. Pink salmon are a major predator and prey species in the PWS ecosystem and provide transport of nutrients

from the marine to terrestrial ecosystem. Pink salmon also support large commercial, sport, and subsistence fisheries which are vital to the economy of the area.

2. Relation to Other Damage Assessment/Restoration Work:

The foundations for this project date back to the original NRDA F/S Study 2 (Injury to Salmon Eggs and Preemergent Fry). NRDA F/S Study 2 was equivalent to the field monitoring portion of this project (Component A) and was conducted in 1989, 1990, and 1991. The same project was continued as Restoration Study R60C in 1992. Two additional elements (Components B and C) were added to Restoration Study R60C during the summer of 1992. These additions were designed to assess the genetic damage hypotheses raised through NRDA F/S Study 2. All three components were present in the 1993 project, Restoration Study 93003. This project, 94191, is the continuation of work started in Restoration Study R60C and continued in Restoration Study 93003.

Several past NRDA and present Restoration projects have been and continue to be intimately related to this project. The 1989 and 1990 NRDA F/S Study 4 demonstrated reduced growth and survival for salmon which reared in oiled areas. NRDA F/S Study 1 in 1989, 1990, and 1991 and subsequent Restoration Study R60B in 1992, investigated oil damage to adult pink salmon spawning populations and provided valuable improvements in escapement estimation procedures used by fisheries managers to monitor and protect injured wild pink salmon populations. NRDA F/S Study 3 in 1989, 1990, and 1991 and subsequent Restoration studies R60A in 1992 and 93185 in 1993 provided hatchery and wild catch contribution estimates. This information was used by fisheries managers to reduce fisheries exploitation rates on injured wild pink salmon and also provided survival estimates for groups of fish examined by NRDA Study 4. The 1989, 1990, and 1991 NRDA F/S Study 28 and a subsequent Restoration study in 1992, incorporated data from all the previous studies into life history and run reconstruction models. These models were used to extrapolate losses in adult pink salmon production from injuries observed in earlier life history stages.

3. Objectives:

a. Component A - Recovery Monitoring of Injury to Pink Salmon Eggs and Preemergent Fry in Prince William Sound

1. Estimate the density, by tide zone, of preemergent fry in 48 streams and eggs in 31 streams using numbers of live and dead eggs and fry.
2. Estimate egg mortality and overwinter survival of pink salmon eggs in both oiled and unoled (control) streams.
3. Assess any loss in adult production from changes in overwinter survival using the results of NRDA F/S Studies 1, 2, 3, and 4.

b. Component B - Verification of Injury to Pink Salmon Gametes in Prince William Sound

1. Determine whether the increased pink salmon egg mortalities observed in oiled streams can be attributed to the physical characteristics of the study streams.

c. Component C - Laboratory Verification of Injury to Pink Salmon Eggs and Preemergent Fry Exposed to Oiled Incubation Substrate.

1. Determine survival, genetic damage, hydrocarbon uptake, mixed function oxidase activity, and sublethal teratogenic effects from long term exposures to oil in eggs exposed from fertilization to emergence.
2. Determine growth characteristics from each exposure group from juvenile stage to maturity.
3. Assess whether differences exist among exposure groups with respect to fecundity, fertilization rate, genetic damage, and sub-lethal teratogenic effects in the second generation progeny through swim-up.

d. Combining Field Observations and Laboratory Results.

1. Determine if the elevated egg mortalities in 1989 and 1990 were potentially caused by oiling in the environment.
2. Determine if the elevated egg mortalities in oiled streams in 1991 were potentially caused by heritable damage to 1989 embryos.

4. Methods:

a. Component A - Recovery Monitoring of Injury to Pink Salmon Eggs and Preemergent Fry in Prince William Sound

1. Data Collection

There are approximately 900 anadromous fish streams in PWS. Preemergent fry sampling from some of these streams has historically provided a pink salmon abundance index which was used to forecast future returns. In recent years, 25 index systems considered representative of pink salmon producing streams have been sampled. Sampling had been performed on as many as 45 streams prior to 1985. This study is designed to compare rates of mortality and abundance among areas with various levels of oil impacts.

Sampling will consist of egg deposition surveys performed from late September to mid-October and preemergent fry sampling conducted from mid-March to mid-April. Streams known to have sustained no oil impact, some oil impact and visibly obvious impact will be included in both the egg and fry sampling programs.

Egg sampling will be conducted in the fall on 31 streams (Figure 1). Fry sampling will be conducted in the spring on 48 streams (Figure 2). These 48 streams will include the 31 streams in the egg sampling program as well as 17 additional streams. The additional streams are those which have traditionally been sampled as part of the historic PWS preemergent index program used to forecast adult returns. Funding for sampling of the preemergent index streams is provided by ADF&G and is independent of this restoration project.

The 31 streams common to the egg and fry sampling programs were selected using the following criteria:

- (1) Adult salmon returns were expected to be great enough to indicate a high probability of success in egg and fry sampling.
- (2) Egg and fry sampling had been done in past years.
- (3) Streams with low to no oil impact, i.e., controls, were selected in the immediate vicinity of high oil impact streams to help account for possible variability in egg and fry survival due to different 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 oil impacts ranging from light to heavy. Most of the 31 selected streams which sustained suspected or obvious oil impact were not sampled for either eggs or fry prior to the EVOS. Among the 12 streams where oil was visibly present in 1989, one had a history of egg sampling and four had a history of fry sampling.

Sampling methods are identical for the preemergent fry and egg sampling and are modeled after procedures described by Pirtle and McCurdy (1977). On each study stream, four zones, three intertidal and one above most tidal influence, will be measured from the mean low tide mark using tide computer generated tide tables and a surveyors level. Boundaries between zones will be marked with stakes. The zones are 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). Separate linear transects 30.5 m in length will be established for egg and preemergent fry samples in each zone (one transect for each type of dig in each zone). The transects will run diagonally across the stream. To insure continuity of transects between egg and fry sampling between years, transect locations are marked with stakes or cairns and carefully photographed from at least two perspectives. To minimize site effects, fall egg and spring fry sampling transects must be located in the same section of stream yet must not overlap, if fall egg sampling is not to influence perceived abundance of fry during spring sampling. To minimize overlap yet allow sampling at the same sites for both eggs and fry, the downstream end of egg sampling transects is located against one bank of the stream and the downstream end of the fry sampling transect is located at the same stream location but against the opposite bank. Fourteen 0.3 m², circular digs (56 per stream) will be systematically made along each transect using

a high pressure hose to flush eggs and fry from the gravel. Eggs and fry will be caught in a specially designed net.

The following data will be collected for each tide zone transect during both egg and fry sampling:

- (1) The sample date.
- (2) The sample tide zone.
- (3) The start and stop time for each tide zone transect.
- (4) Numbers and condition (live or dead) of fry and eggs by species for each dig.
- (5) A subjective estimate of the overall percent yolk sac absorption for fry in each dig sample.

Data will be entered from "Rite in the Rain" books into a Lotus spreadsheet for editing and summarization.

Pink salmon eggs will be separated from chum *O. keta* and coho *O. kisutch* salmon eggs by their smaller size. Chum salmon eggs will be separated from coho salmon eggs by their greater development and different coloration. An egg will be considered dead if it is opaque or discolored with concentrations of lipids. Pink salmon fry will be differentiated from chum salmon fry by their smaller size and lack of parr marks. Sampling will often kill fry (especially newly hatched fry), so fry will only be considered dead if decomposition is evident.

2. Data Analysis

Numbers of live and dead preemergent fry and eggs will be summarized by date, stream, level of hydrocarbon impact, and stream zone. Densities of live eggs for stream i , zone j in m^2 (E_{ij}) will be estimated by:

$$\hat{E}_{ij} = \frac{\sum LE_{ijk}}{0.3n_{ij}} \quad (1)$$

where LE_{ijk} is the number of live eggs found in the k^{th} dig, in stream i , zone j , and n_{ij} is the number of digs from stream i , zone j . Densities of dead eggs as well as dead and live fry will be calculated using the same estimator with appropriate substitutions.

Pink salmon egg mortality will be estimated for each stream using the following relationship:

$$\hat{M}_{ij} = \frac{\Sigma(DE_{eijk} + DF_{eijk})}{\Sigma(LE_{eijk} + DE_{eijk} + LF_{eijk} + DF_{eijk})} \quad (2)$$

where DE_{eijk} , DF_{eijk} , LE_{eijk} , and LF_{eijk} are the number of dead eggs, dead fry, live eggs, and live fry for the k^{th} dig from stream i , zone j , collected during egg dig e , respectively.

The Arcsin square root transformation will be examined as well as the Logit transform of egg mortality [$\ln(\text{odds})$].

$$\text{Logit}_{ij} = \ln \left[\frac{\Sigma(DE_{eijk} + DF_{eijk})}{\Sigma(LE_{eijk} + LF_{eijk})} \right] \quad (3)$$

Pink salmon egg to preemergent fry survival will be estimated as:

$$\hat{S}_{ij} = \frac{(\Sigma LF_{fijk}) / n_f}{\Sigma(LE_{eijk} + DE_{eijk} + LF_{eijk} + DF_{eijk}) / n_e} \quad (4)$$

where LF_{fijk} is the number of live fry for the k^{th} dig f from stream i , zone j , collected during fry dig f , and n_e and n_f are the number of digs for stream i , zone j for egg dig e and fry dig f .

Differences in egg mortality and survival will be examined using a mixed effects two-factor experiment with repeated measures on one factor (Neter et al. 1990):

$$Y_{ijk} = \mu_{...} + O_i + Z_j + (OZ)_{ij} + S_{k(i)} + e_{(ijk)} \quad (5)$$

The two treatments will be extent of oiling, (O_i , 2 levels; oiled and unoled), and height in the intertidal zone (Z_j , 4 levels; 2.1, 2.7, and 3.4 m above mean low water, and upstream) both fixed effects. The data will be blocked by stream ($S_{k(i)}$), a random effect nested within 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, 1969), while normality will be visually assessed using normal quantile-quantile and box plots (Chambers et al. 1983). If the data appear to be non-normal, data transformations will be examined. If a significant difference due to oiling is detected ($\alpha = 0.05$), four contrasts (oil vs.

unoiled 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 the hydrocarbon results from mussel samples (NRDA F/S Study 1). Different groupings of oiled and unoiled streams will be analyzed, if evidence of oiling is not consistent.

b. Component B - Verification of Injury to Pink Salmon Gametes in Prince William Sound

1. Experimental Design

The experiment will assess the effects of the physical characteristics of the study streams upon the observed results. This will be accomplished by collecting pink salmon gametes from oiled and non-oiled streams and rearing the resulting embryos in a controlled laboratory environment.

This experiment will provide information to help determine whether the results observed in NRDA Study FS2 can be attributed solely to the physical characteristics of the study streams. In this experiment we will collect gametes from 8 oiled and 8 non-oiled streams from southwestern PWS, make intra-stream crosses, and incubate the resulting embryos in a controlled laboratory environment. Egg mortality will be compared between the oiled and uncontaminated streams. If no difference is observed in this experiment and a significant difference in egg mortality is detected between oiled and non-oiled streams during the recovery monitoring portion of this study during the fall of 1993 egg sampling, it can be stated that the physical characteristics of the study streams played a role in the results of the previous egg mortality studies.

Gamete collection and fertilization procedures will occur over a four day period to obtain data from 8 oiled and 8 non-oiled streams. Gametes from 30 male and 30 female pink salmon will be collected from 2 oiled and 2 control streams during each sampling day. The gametes will be flown to the Armin F. Koernig hatchery where a random gamete pool will be assembled for each stream in a timely manner. The construction of the random gamete pool is described in the fish culture section of this proposal. 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 (Heath Incubator).

Incubating embryos will be periodically screened for dead eggs and hatching success. Samples of sperm from each male used to build the embryo pools will be cryopreserved for future analysis if required. Embryo samples will also be collected and preserved for future examination by flow cytometry, MFO, and histopathology. The experiment will be terminated prior to swimup at which time all larvae will be killed.

2. Data Analysis

The data will be analyzed as a fixed-effects generalized randomized block design:

$$Y_{ijk} = \mu + B_i + O_j + \epsilon_{ijk} \quad (6)$$

where Y_{ijk} is egg mortality for sample day i , oil contamination level j , and stream k ; μ 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 ϵ_{ijk} is random error. The relative power of the test was estimated. The sample size was considered sufficient to detect a difference of less than 1.5 standard deviations at $\alpha = 0.05$ and 95% power (Neter et al. 1990). A test with high power is needed to protect against arriving at the conclusion that all observed damages could be attributed to the physical characteristics of the streams when in actuality significant damages due to oil were present.

The assumption of constant error terms will be tested using the F_{\max} -test (Sokal and Rohlf 1969) 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. Egg fertilization and incubation

Gametes will be randomized as described below, and embryos will be incubated in Heath incubators located at the Armin F. Koernig hatchery in Prince William Sound. Each incubator tray will have an independent water supply from a common water source.

c. Component C - Laboratory Verification of Injury to Pink Salmon Eggs and Preemergent Fry Exposed to Oiled Incubation Substrate.

1. Experimental Design

This experiment measures differences in biological response to various concentrations of Exxon Valdez crude oil over two brood years. It will be a controlled simulation, incorporating our observations of field conditions. This study will span two generations in order to evaluate the findings of Sharr et al. (1994a, 1994b, and in prep) and Bue et al. (in press). The first generation will verify the 1989 and 1990 findings while the second generation will provide evidence to confirm the functional sterility hypothesis. This study will also provide samples of known oiling history for examination of genetic material through the use of flow cytometry.

Fertilized eggs will be incubated on substrates treated with different levels of oil and reared to emergence. Biological responses to the oiled substrate will be evaluated. Surviving fry will be cultured to maturity at which time their gametes will be collected, crossed, and incubated in a clean environment. Differences in gamete fertilization rates and embryo survival will be compared with the oil exposures experienced by the parental generation. Two brood years (1992 and 1993) will be examined to ensure success of the experiment.

Brood Year 1992

The brood year 1992 experiment will examine the effects of six levels of intertidal gravel oil contamination (five oiled and a control) on responses to various life history stages of pink salmon across two generations. Responses to be measured in the first generation include survival to eyeing, survival to emergence, hydrocarbon uptake, and size at emergence. The first generation will reach maturity in September 1994. At that time, survival, growth to maturity, and fecundity will be observed. Responses measured in the second generation will include fertilization rate, survival to each major developmental stage, and number of defective progeny at emergence.

Gametes from 48 male and 48 female pink salmon will be collected, randomly mixed into a common embryo pool, and divided into 48 aliquots of approximately 1500 eggs each. The aliquots will be randomly assigned to one of the six gravel treatments (8 aliquots per treatment). The aliquots will be incubated in individual pipe incubators filled with treatment gravel. Sampling will take place at each major developmental stage; eyeing, hatching and emergence. Samples will be randomly removed from the incubators for genetic, mixed-function oxidase (MFO), histopathological, and hydrocarbon analysis. Fry will be counted and inspected upon emergence and then moved to saltwater netpens. Water samples collected in conjunction with the embryos will be used to establish oil dosages in each incubator. Intra-group pairings will be made for each of the four remaining first generation treatment groups. Confining the experiment to within group pairings simulates the natural homing characteristics of pink salmon and the relatively low levels of genetic interchange thought to occur between streams in the wild. Second generation pairings will again use a randomly mixed common gamete pool utilizing equal numbers of males and females. These gametes will not be incubated in an oiled environment hence any observed increases in mortalities or defective individuals can be attributed to oiling effects upon the first generation. These eggs will be incubated through emergence. Number of defective progeny will be compared between treatment groups.

Brood Year 1993

This experiment will be repeated for the 1993 brood year using an additional two levels of oiled gravel (eight levels in all).

2. Data Analysis

The data from each generation will be analyzed as a fixed-effects factorial design:

$$Y_{ij} = \mu + C_i + \epsilon_{ij} \quad (7)$$

where Y_{ij} is the j^{th} response to oiling concentration i , μ is the model mean, C_i is the level of oil concentration (six levels-five oiled and a control), and ϵ_{ij} is random error. The power of this test was estimated using data from past pink salmon incubation studies (Wertheimer 1985). These data indicated the ability to detect a difference of less than 10% in survival to emergence at $\alpha = 0.05$, 90% of the time.

The assumption of constant error terms will be tested for all analysis using the F_{max} -test (Sokal and Rohlf, 1969) 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 contrasts will be made using Bonferroni family confidence intervals. The SAS (SAS Institute Inc., 1981) General Linear Models Procedure will be used to analyze the data.

3. Fish Culture

All experiments in component C will be performed at The National Marine Fisheries Research Station at Little Port Walter (LPW) in southeastern Alaska. Mature pink salmon gametes will be collected from intertidal spawners in Lover's Cove Creek located near the facility.

a. Randomization of Gamete Pools

The randomized embryo pool will be created by (1) spawning the females into a common container, (2) randomizing the eggs within the container, (3) dividing the eggs into aliquots, (4) fertilizing each aliquot with an individual male, and (5) again recombining all fertilized aliquots into a composite embryo pool. The aliquots used in the experiment will then be randomly drawn from the composite embryo pool.

b. Incubation

Pipe incubators will be used to simulate in stream incubation. These incubators will be constructed from 30 cm sections of 16 cm polyvinylchloride pipe. The pipe will be stood on end, sealed, and fitted with a water intake at the bottom. The pipe will then be filled with appropriately treated gravel. This design allows water to upwell through the gravel and out an outlet fitting at the top of the incubation pipe.

Fertilized eggs will be laid on top of the gravel to incubate. Upon hatching, the alevins will be permitted to burrow into the substrate. Eggs will be exposed to

saltwater for 4 hour intervals every 12 hours during incubation to simulate intertidal incubation. Emerging fry will be removed to saltwater netpens.

c. Culture to maturity

All fry will be raised to maturity using standard hatchery procedures. They will be fed a commercial diet, vaccinated against *Vibrio anguillarum*, and treated with antibiotics as needed. Maturing fish will be fed a commercially available brood diet.

The treatment groups will be reared in separate netpens until they are 6 g at which time they will be tagged with passively induced transponders (PIT tags). PIT tags provide individual fish with unique identification codes which can be interrogated without harming the fish. Approximately 300 fish from each treatment group will be tagged. Each set of tagged fish will be split into two equal size groups and placed into one of two netpens. Each netpen will contain fish from all treatment groups. One netpen will be kept at LPW while the other will be maintained 5 km to the north at Osprey Bay to ensure survival of the experiment. Fish will be counted and measured for length and weight each fall and spring to establish survival and growth rates during the experiment.

4. Development of Dose Response Curves

Dosing levels in Studies 1 and 2 of Component C will be established by analyzing hydrocarbon concentrations in incubator effluent and food with gas chromatograph and mass spectroscopy (GC/MS) at each major developmental stage. Effluent samples for the GC/MS will be collected and pooled from each of the pipe incubators in an oiling concentration-duration of exposure treatment. It will not always be necessary to sample all of the treatment cells in the experimental design as the number of uniquely exposed treatment groups changes with embryo development. For example, at eyeing there are 6 uniquely exposed groups since all exposures have been made for the same amount of time at 6 different oil concentrations; however, at emergence there are 11 uniquely exposed treatment groups, different concentrations have been applied over 2 different durations. Additional effluent samples will be collected at each major developmental stage for spectrophotofluoremetry to provide estimates of variability between incubators within a treatment cell. Oil concentrations in incubator gravel will be obtained from spectrophotofluoremetry and related to levels observed in streams sampled under NRDA. Each treatment cell with a unique exposure level will be sampled at least 3 times for tissue hydrocarbon concentration. Samples will be collected at all stages from eyeing to 6 weeks after emergence.

d. Flow Cytometry

Flow cytometry will be used to analyze the DNA content of whole embryos and individual tissues (e.g., liver, kidney, gonad, gill) as called for at the appropriate test points in experiments performed under components B and C (e.g., Kocan and Powell 1985, McBee

and Bickham 1988). All analyses will be made on fresh tissues prepared no more than 24 hours prior to flow cytometry analysis.

Suspensions of stained nuclei will be produced for DNA content analysis using nuclear isolation medium (NIM) (0.9% NaCl, 10 mM Tris, 2 mM CaCl_2 , 2 mM MgCl_2 , 0.1% Nonidet P-40, 106 mM MgSO_4 , and 1 mg/100ml DAPI (4,6-diamidino-2-phenylindole dihydrochloride)) (e.g., Thornthwaite et al., 1980, and Seeb et al. 1988). Embryos and tissue samples will be placed into 1.5 ml microcentrifuge tubes containing 1 ml of NIM. Samples will be minced in 0.5 ml of NIM approximately one minute using two scalpels to obtain a cellular suspension, allowed to incubate at $2-3^\circ\text{C}$ for 15 min, and filtered through a $40\ \mu\text{m}$ nitex nylon filter to remove debris and clumped cells. Stained nuclear suspensions will be refrigerated overnight for flow cytometry analysis the following day. Immediately prior to flow cytometry analysis samples will be triturated 3 times using a 26 g syringe and filtered through $40\ \mu\text{m}$ nitex nylon filter to remove any residual clumps of nuclei. Samples will be analyzed using a PARTEC PAS II flow cytometer with optical filters for DAPI excitation and ACQCYTE data acquisition and MULTICYCLE DNA analysis software (Phoenix Flow Systems Inc. 1991) following the methods of Lamb et al. (1991).

e. Alternatives

Several short-term cytogenetic assays exist in addition to flow cytometry for evaluating the potential genotoxic effects of chemicals and compounds. These methods are designed to identify four general types of genetic changes: DNA microlesions and macrolesions, primary DNA damage, and morphologic changes in target cells (Brusick 1987). Of these, assays for the detection of DNA macrolesions and primary DNA damage are generally accepted as being standard for identifying genotoxic agents.

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). These techniques are capable of detecting and quantifying subtle chromosome changes. However, these techniques have disadvantages as well. Physical separation of metaphase and anaphase chromosomes for visual scoring is required. The techniques for chromosome separation and isolation can be technically involved and are not standardized between laboratories. Visual scoring of the desired endpoints can be somewhat subjective. The time involved for isolating and scoring chromosomes limits sample sizes to 100-200 cells which reduces statistical accuracy and precision.

The need for large sample sizes cannot be solved by conventional cytogenetic techniques and has been the motivating force behind development of flow cytometry for cytogenetic testing (Deaven 1982). Flow cytometry allows analysis of large numbers of cells (10^3 - 10^5) greatly increasing statistical power. 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). 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.

Flow cytometry analysis provides a more comprehensive measure of genetic damage than traditional cytogenetic techniques and 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). Once flow cytometry has demonstrated the presence of genetic damage it may be useful to apply traditional cytogenetic techniques on a more limited scale to identify specifically what type of damage has occurred.

An additional procedure for detecting deleterious mutations that has just been brought to our attention utilizes a technique termed androgenesis. Androgenetic individuals are obtained by enucleating eggs with gamma radiation before fertilization; fertilization is then done with normal sperm. If no other treatments are applied, the resulting progeny will be haploid, containing only a single set of chromosomes from the male parent and none from the female. Mortality rates for these haploids are directly related to the presence and number of deleterious mutations (Armstrong and Fletcher, 1983). Advantages of this technique over most 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 by laboratories because of the requirement of a gamma radiation treatment. Yet it is thought to be one of the most sensitive techniques available to detect the presence and effects of deleterious recessive mutations. We propose to utilize the expertise of a contractor to perform a pilot study. Haploid androgens will be used to test for the presence of deleterious mutations in the chromosomes of populations still suffering increased rates of mortality.

5. Location:

Component A: Spring fry sampling will be conducted on 48 streams (Figure 1). These will include the 25 streams in the ongoing ADFG preemergent index program plus 23 additional streams. The additional streams are located in Central and Southwest PWS where most of the oiling occurred. Egg sampling will be conducted in the fall on 31 of the 48 streams sampled for preemergent fry (Figure 2). Streams included in the fry sampling program but not in the egg program are traditional fry sampling streams located on the eastern and northern shore of PWS. These streams are outside the area studied for oil impact effects.

Component B: The experiment designed to evaluate the effects of environment on egg mortality will collect gametes from streams in Western Prince William Sound and incubate the resulting embryos at the Armin F. Koernig hatchery in Southwestern Prince William Sound (Figure 3).

Component C: The experiments designed to test the effects of oiled incubation substrate on gamete viability will be performed at the National Marine Fisheries Service Laboratory at Little

Port Walter, Baranof Island, southeastern Alaska (Figure 4). Hydrocarbon analysis will be performed at the National Marine Fisheries Service Auke Bay Laboratory.

All work dealing with the assessment of genetic damage will be performed at the Regional Fish and Game Office in Anchorage.

6. Technical Support:

A biometrician will ensure the study design will provide a reasonable chance of reaching a defensible conclusion.

Flow cytometry specialist will ensure proper tissue collection and preparation procedures, operate the flow cytometer, and assist in histogram interpretation and analysis.

A chemist is required to establish a dosing protocol, determine hydrocarbon concentrations, and evaluate results of hydrocarbon analysis.

7. Contracts:

Sole source contracts will be required with the University of California, Davis and the Woods Hole Oceanographic Institute for histopathological and mixed-function oxidase work. These institutions have done the majority of this type of work under damage assessment and it is essential that the results of this controlled experimentation be consistent with the results gathered under NRDA.

We propose to have a \$15,000 project *Detection of Deleterious Mutations in Pink Salmon Through Haploid Androgenesis* conducted by Dr. Gary Thorgaard, Washington State University (WSU), as a sole-source contractor (see attached study design). 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. Dr. Thorgaard's proposal is attached.

D. SCHEDULES

COMPONENT A - Recovery Monitoring of Injury to Pink Salmon Eggs and Preemergent Fry in Prince William Sound

Dates	Activity
1 Oct - 15 Oct 1993	Egg deposition sampling.
30 Oct 1993 - 30 Jan 1994	Analysis of 1993 egg data and completion of first draft of 93003 report for egg and fry data
7 Mar - 10 Apr 1994	Preemergent fry sampling on 48 streams.
1 May - 1 Sep 1994	Analysis and preliminary summarization of 1994 preemergent fry data.
15 Sep - 15 Oct 1994	Egg deposition sampling.
30 Oct 1993 - 30 Jan 1995	Analysis of egg data and completion of first draft of 94191 report for egg and fry data.

COMPONENT B - Laboratory Verification of Injury to Pink Salmon Eggs and Preemergent Fry in Prince William Sound

Dates	Activity
30 Oct 1993 - 30 Jan 1994	Analysis of 1993 data and completion of first draft of 93003 report for laboratory evaluation
1 Aug - 15 Aug 1994	Preparation for Experiment
15 Aug - 30 Aug 1994	Collect Gametes and make crosses from 16 streams
30 Aug - 15 Nov 1994	Monitor incubators and collect data
15 Nov 1994 - 30 Jan 1995	analyze data and prepare first draft of 94191 report for laboratory evaluation

COMPONENT C - Laboratory Verification of Injury to Pink Salmon Eggs and Preemergent Fry in Prince William Sound

1992 Brood Year

Period	Complete	Tasks
Jul 15 - Sep 15 1992	X	Oil gravel, set up incubators
Sep 15 1992 - Sep 15 1993	X	Spawn pink salmon, collect incubation data, pond fry, culture fry, PIT tag and move to netpens.
Sep 15, 1993	X	Write first interim report
Sep 15, 1993 - Sep 15 1994		Culture tagged fish in netpens, observe growth rates, size at maturity and fecundity. Obtain gametes, spawn second generation.
Sep 15 1994		Write second interim report
Sep 15 1994 - May 15 1995		Incubate second generation, observe survival to each major developmental stage.
May 15 - Aug 15 1995		Analyze and integrate data collected from culture of 1992 pink salmon and their progeny.
Sep 15 1995		Write third interim report

1993 Brood Year

Period	Complete	Tasks
Jul 15 - Sep 15 1993	X	Oil gravel, set up incubators
Sep 15 1992 - Sep 15 1994		Spawn pink salmon, collect incubation data, pond fry, culture fry, PIT tag and move to netpens.
Sep 15, 1994		Write second interim report
Sep 15, 1994 - Sep 15 1995		Culture tagged fish in netpens, observe growth rates, size at maturity and fecundity. Obtain gametes, spawn second generation.
Sep 15 1995		Write third interim report
Sep 15 1995 - May 15 1996		Incubate second generation, observe survival to each major developmental stage.
May 15 - Aug 15 1996		Analyze and integrate data collected from culture of 1993 pink salmon and their progeny.
Aug 15 - Sep 15 1996		Write final report

E. EXISTING AGENCY PROGRAM

An additional 30.0 K will be provided by the Alaska Department of Fish and Game through normal operating funds. This amount is budgeted to cover the normal preemergent fry sampling program which has been conducted annually since 1961.

F. ENVIRONMENTAL COMPLIANCE/PERMIT/COORDINATION STATUS

Egg and preemergent fry sampling will require an ADFG Title 16 permit and an ADFG biological collections permit. Transport of wild gametes to the PWSAC hatchery on Evans Island, PWS will require an ADFG Fish Transport Permit for each stock and a permit. Alteration may be required to rear and incubate the wild eggs at the hatchery.

An ADFG Fish Transport Permit will be required to obtain broodstock from Lover's Creek for the Laboratory verification of Injury work at Little Port Walter.

PERFORMANCE MONITORING

This will be a joint project between ADFG and NMFS. ADFG will be the lead agency for overall program management and genetic damage determinations. ADFG will be responsible for data collection, gamete fertilization, and incubation in Components A and B. NMFS will be responsible for the oil exposures, chemistries, fish culture, and hydrocarbon end points in Component C. Both agencies will have statistical analyses responsibilities, particularly with the experimental designs. Both agencies will have joint responsibilities for meshing the lab and field results to reach a conclusion in the study.

For ADFG, principal investigator Sharr (Fisheries Biologist III) and his assistant Sharp (Fisheries Biologist II) will provide field results to date, help design the laboratory experiment, and insure that laboratory conditions and treatments simulate those observed in wild streams. Principal investigator Seeb (Principal Geneticist) will help design and provide genetics oversight for the laboratory rearing of wild embryos as well as the flow cytometry portions of the experiment. He will also supervise the collection and analysis of flow cytometry samples. Consulting biometrician Bue (Biometrician II) will conduct the experimental design and provide statistical oversight for the project. Sharr, Seeb, and Bue will cooperate in the data analysis and writing of project reports.

Most methods to be incorporated in the ADFG portions of this project have been used before, some for many years, all are now standardized and well documented in operational plans. ADFG project personnel including most of the project technicians have participated in sampling activities and laboratory rearing activities associated with Component A and portions of Component B. Persons supervising field sampling in Component A receive annual training at one or more area hatcheries with respect to speciating eggs and fry and making live and dead determinations. The principal geneticist and his staff have extensive laboratory fish culture experience and will be present at all times during the rearing experiment at AFK Hatchery. One member of the permanent Cordova ADFG staff has been an assistant principal investigator for this project in the past and could be called upon to temporarily resume those duties should the need arise. Additionally, several other members of the Cordova ADFG staff have participated in field sampling and aquaculture portions of the project in 1992 and prior years and could be called upon in case of personnel shortages.

For NMFS, overall supervision of this project will rest with NMFS GS-14 physiologist, principal investigator (Rice). The PI will be responsible for monitoring the progress of the project, provide quality control for the design and implementation, oversee the budget and review all interpretations in the products. In addition, the PI will supervise two primary task leaders: a GS-11 biologist (Heintz) assigned to LPW, and a GS-13 chemist (Short). The GS-11 biologist will direct field sampling and data collection, fish culture, and perform statistical analysis of the data. A GS-9 biologist will assist the GS-11 biologist in setting up the experiment and collecting data. Technicians will be required to perform detailed fish culture such as incubator maintenance and fish feeding. The GS-13 chemist is responsible for developing dosing techniques and analyzing samples for the presence of hydrocarbons, and interpreting the hydrocarbon analyses.

Data will be recorded in an Rbase database. There will be several data tables in the database, including "incubation", "rearing" and "spawning". The incubation table will include incubator number, number eggs seeded into incubator, and for each developmental stage: water chemistry, hydrocarbon concentrations, MFO presence, coefficient of variation for cellular DNA content, and number surviving to emergence. The key field that links the "rearing" table with the "incubation" table will be incubator number. The "rearing" table will also include PIT tag code, length and weight at each sample point. The "spawning" table will include the first generation incubator number, second generation incubator number, second generation fertilization rate, first generation fecundity, survival to eyeing, hatching, and emergence.

Graphical summaries of data will be made using LOTUS 123, and statistical analysis will use SAS and MINITAB. All raw and summarized data and reports are stored as hard copy and electronically on diskettes in two separate locations at the NMFS Auke Bay Lab. Quality assurance and documentation of all databases structures will be reviewed by FS 30 (Database Management) personnel in Juneau and duplicates of all database documentation will be maintained in their files.

Biological samples for hydrocarbon, MFO, and DNA analyses will be clearly labelled both on the inside and outside of the container with indelible ink. Samples will be stored in freezers at the NMFS Auke Bay Lab.

Field activities will continue until injury to salmon eggs 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 egg and preemergent fry data. The most significant information on damages demonstrated in 1989 through 1991 will be written up as a close out report for the NRDA Study and will also be published in a juried 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.

H. COORDINATION OF INTEGRATED RESEARCH EFFORT

The field data collection for Component A of this project is very specific to individual wild pink salmon streams and precedes or follows most field activities of SEA (94320) 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 Study and these data will be utilized by several SEA studies.

Data collection for Component B occurs over a very compressed period of time and is again very stream specific hence does not blend well logistically with most of the other pink salmon studies. However, all of the streams used for brood stock in this study are also of interest to the Pink Salmon Genetics Project (94189) and carcasses from the egg takes can be used for genetic sampled from these streams. The study is housed at the PWSAC AFK Hatchery and will take advantage of the same incubation facilities as the PWSAC experimental release project included in the SEA project.

Component C of this project occurs at Little Port Walter on Baranof Island in Southeast Alaska and there is little opportunity for coordination with other projects with respect to data collection.

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.

I. PUBLIC PROCESS

Many of the field procedures used in the field monitoring portion (Component A) of this project have been employed as part of the data collection activities for preemergent fry indices used in PWS pink salmon forecasts for more than 30 years. The procedures have been presented and reviewed at a multitude of workshops and scientific meetings, are widely understood by the fishing industry, and have undergone peer review through the NRDA process. Field monitoring methodologies were presented at the 1991 Pink and Chum Workshop in Parksville, British Columbia, Canada. Field monitoring results from 1989, 1990, 1991, and 1992 were presented at the 1993 meeting of the Alaska Chapter of The American Fisheries Society in Valdez, Alaska, the 1993 Oil Spill Symposium in Anchorage, Alaska, and the 1993 Pink and Chum Workshop in Juneau, Alaska. Abbreviated operational plans for 1989 through 1994 egg and alevin mortality studies have been published annually in EVOS Trustee Council work plans which incorporate public comment.

J. PERSONNEL QUALIFICATIONS

Fisheries Biologist III - Samuel Sharr

Mr. Sharr received a Bachelor of Science degree in biology from the University of Washington in 1968. He has been a research biologist for ADFG since 1979 and has worked on PWS salmon and herring since 1981. He assumed his present position as the ADFG, Division of Commercial Fisheries, Biologist III, PWS Area Finfish Research Project Leader in 1986. In this capacity, Mr. Sharr oversees all the salmon and herring research conducted by the Division of Commercial Fisheries in PWS. His involvement with the PWS salmon escapement aerial survey program dates from the early 1980's. Mr. Sharr has supervised a total re-edit of the historic aerial and ground survey data and designed a new RBASE data base for inseason escapement analyses. Mr. Sharr wrote the original operational plans for NRDA F/S Studies 1,2 and, 3 and has been the Principal Investigator for those projects since their inception.

Principal Geneticist - James E. Seeb

Jim Seeb earned a B.S. in Biology (1974) from the University of Puget Sound, an M.S. in Fisheries (1982) and a Ph.D. in Fisheries (1987) from the University of Washington. Jim has worked as a Fish Biologist for the Washington Department of Fisheries (1978-1980) and Pacific Fisheries Research (1980-1982), as a Graduate Research Assistant at the University of

Washington (1982-1986), a Research Assistant Professor at the University of Idaho (1987-1988), and as an Assistant Professor at Southern Illinois University (1988-1990). Presently, Jim is the Principal Geneticist for FRED Division of the Alaska Department of Fish and Game and has overall responsibility for fisheries genetic issues throughout Alaska. Dr. Seeb has published extensively in the Fisheries and Genetics Literature. He has worked with many fish species on numerous genetic topics including but not limited to genetic marking and its use to assess stock dynamics and management programs, genetic variation and postglacial dispersal of populations, the use of genetic structure in the enforcement of fishing regulations, and the measurement of DNA content using flow cytometry.

GS-14 Physiologist - Stanley D. Rice

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

Biometrician II - Brian G. Bue

Brian Bue has a Bachelor of Science in Biology and a Bachelor of Science in Fisheries from the University of Alaska, Fairbanks. He also possesses a Masters degree in Fisheries with an emphasis on quantitative studies from the University of Alaska, Fairbanks. Brian has worked with the Alaska Department of Fish and Game from 1974 through present in many capacities. He has worked as a consulting biometrician on oil spill damage assessment projects since the first days of the *Exxon Valdez* spill.

GS-13 Chemist - Jeffrey Short

Mr. Short is an analytical chemist at the Auke Bay Laboratory (ABL), and leads the hydrocarbon analysis facility at ABL, which is one of the two laboratories analyzing *Exxon Valdez* NRDA hydrocarbon samples. Mr. Short holds a B.S. in biochemistry and an M.S. in physical chemistry from the University of California. He is principal investigator (PI) of NRDA project Subtidal Study #3, and was among the first scientists to collect samples 7 days after the spill: he was awarded both individual and unit citations from NOAA for these efforts. Mr.

Short has conducted extensive research on the effects of Alaskan crude oils to Alaskan marine biota over a period of 10 years prior to the *Exxon Valdez* oil spill.

GS-11 Fisheries Biologist (Research) - Ron A. Heintz

Ron Heintz has a Bachelor of Science in Ecology from the University of Illinois (1979), and a Masters degree in Fisheries from the University of Alaska, Fairbanks (1987). He has worked for the National Marine Fisheries Service since 1985 concentrating his efforts on salmon enhancement research. He is the principal investigator and co-investigator on several salmon genetics projects.

Fisheries Biologist II - Gary Miller

Gary Miller is the flow cytometry specialist for the Alaska Department of Fish and Game Genetics Laboratory in Anchorage. Gary has a Bachelor of Science in Fisheries Biology from the University of Washington, a M.S. in Zoology from Southern Illinois University - Carbondale, and is currently pursuing his Ph.D. from the University of Washington. He has worked periodically for the Alaska Department of Fish and Game since 1981. He has a strong background in genetics and developmental biology and has conducted research and co-authored projects in hybridization, polyploid induction, allozyme expression, and growth performance of triploid salmonids and other fishes. He has extensive laboratory experience with techniques including flow cytometry, protein starch gel electrophoresis, protein and molecular marker analysis, and fluorescent antibody testing of pathogens.

K. BUDGET

(see attached)

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 1994 Federal Fiscal Year Project Budget
 October 1, 1993 - September 30, 1994

Project Description: Oil Related Egg & Alevin Mortalities - This project will measure egg and alevin mortalities in oiled and unoled streams and monitor recovery (continuation of 93003). Laboratory rearing and dose response experiments will be conducted to verify oil as the cause for increased mortality observed in oiled streams in 1989 through 1992. These experiments will also examine the possibility of genetic injury as an explanation for chronic injury and assess the likely time frame for natural recovery.

Budget Category:	1993 Project No. 93003 Authorized FFY 9	'93 Report/ '94 Interim* FFY 94	Remaining Cost** FFY 94	Total FFY 94	FFY 95	Comment
Personnel	\$434.2	\$236.7	\$280.8	\$517.5	\$464.1	
Travel	\$39.6	\$12.3	\$23.5	\$35.8	\$32.7	
Contractual	\$184.9	\$36.5	\$109.3	\$145.8	\$94.1	
Commodities	\$86.5	\$26.2	\$43.3	\$69.5	\$67.0	
Equipment	\$88.1	\$2.1	\$15.0	\$17.1	\$7.1	
Capital Outlay	\$0.0	\$0.0	\$0.0	\$0.0	\$0.0	
Subtotal	\$833.3	\$313.8	\$471.9	\$785.7	\$665.0	
General Administration	\$78.0	\$38.1	\$49.8	\$87.8	\$76.2	
Project Total	\$911.3	\$351.9	\$521.7	\$873.5	\$741.2	
Full-time Equivalents (FTE)	9.7	4.4	5.0	9.4	9.4	
Dollar amounts are shown in thousands of dollars.						
Budget Year Proposed Personnel:		Reprt/Intrm Months	Reprt/Intrm Cost	Remaining Months	Remaining Cost	
Position Description						
See Individual 3A Forms for Personnel Details						
Personnel Total		0.0	\$0.0	0.0	\$0.0	
NEPA Cost:						\$0.0
*Oct 1, 1993 - Jan 31, 1994						
**Feb 1, 1994 - Sep 30, 1994						

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Project Number: 94191
 Project Title: Oil Related Egg & Alevin Mortalities
 Agency: AK Dept. of Fish & Game

**FORM 2A
 PROJECT
 DETAIL**

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EXXON VALDEZ TRUSTEE COUNCIL
 1994 Federal Fiscal Year Project Budget
 October 1, 1993 - September 30, 1994

Project Description: Oil Related Egg & Alevin Mortalities - This project will measure egg and alevin mortalities in oiled and unoled streams and monitor recovery (continuation of 93003). Laboratory rearing and dose response experiments will be conducted to verify oil as the cause for increased mortality observed in oiled streams in 1989 through 1992. These experiments will also examine the possibility of genetic injury as an explanation for chronic injury and assess the likely time frame for natural recovery.

Budget Category:	1993 Project No. 93003 Authorized FFY 9	'93 Report/ '94 Interim* FFY 94	Remaining Cost** FFY 94	Total FFY 94	FFY 95	Comment		
Personnel	\$305.8	\$112.0	\$191.8	\$303.8	\$249.1	Report: \$108.4	Rem FY9 \$44.7	FY95 \$21.7
Travel	\$28.6	\$7.1	\$7.3	\$14.4	\$12.7	\$4.9	\$2.0	\$1.8
Contractual	\$68.9	\$36.5	\$59.3	\$95.8	\$69.1	\$0.0	\$22.7	\$1.0
Commodities	\$30.5	\$13.5	\$17.5	\$31.0	\$27.0	\$9.5	\$4.0	\$0.5
Equipment	\$69.1	\$2.1	\$0.0	\$2.1	\$2.1	\$0.0	\$0.0	\$0.0
Capital Outlay	\$0.0	\$0.0	\$0.0	\$0.0	\$0.0	\$0.0	\$0.0	\$0.0
Subtotal	\$502.9	\$171.2	\$275.9	\$447.1	\$360.0	\$122.8	\$73.4	\$25.0
General Administration	\$50.6	\$19.4	\$32.9	\$52.3	\$42.2	\$16.3	\$8.3	\$3.3
Project Total	\$553.5	\$190.6	\$308.8	\$499.4	\$402.2	\$139.1	\$81.7	\$28.3
Full-time Equivalents (FTE)	6.9	1.9	3.1	5.0	5.0		0.7	0.2
Dollar amounts are shown in thousands of dollars.								
Budget Year Proposed Personnel:		Reprt/Intm	Reprt/Intm	Remaining	Remaining	Rem. FY94 and FY95 budgets presented above reflect the additional cost of repeating the pink salmon embryo incubation experiment at the AFK hatchery for the 1994 brood year.		
Position Description		Months	Cost	Months	Cost			
Reprt Fisheries Biologist III		1.5	\$9.9	2.5	\$16.5			
& Fisheries Biologist II (2)		8.5	\$42.5	15.7	\$78.0			
Intm Fisheries Biologist I		1.5	\$6.7	1.2	\$6.0			
Fisheries Tech. III (2)		4.5	\$15.6	8.5	\$32.2			
Fisheries Tech. II (4)		2.4	\$13.0	4.2	\$25.6			
Biometrician I & II		3.0	\$17.6	4.5	\$26.8			
Program Manager		1.0	\$6.7	1.0	\$6.7			
Personnel Total		22.4	\$112.0	37.6	\$191.8	NEPA Cost: \$0.0		
						*Oct 1, 1993 - Jan 31, 1994		
						**Feb 1, 1994 - Sep 30, 1994		

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Project Number: 94191
 Project Title: Oil Related Egg & Alevin Mortalities
 Sub-Project:
 Agency: AK Dept. of Fish & Game

FORM 3A
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EXXON VALDEZ STEE COUNCIL
1994 Federal Fiscal Year Project Budget
October 1, 1993 - September 30, 1994

Travel:		Reprt/Intr	Remaining
Reprt	5 Trips Cordova/Anchorage @ \$500/trip	\$2.5	\$0.0
	4 RT Anchorage/Juneau @ \$600/trip	\$2.4	\$0.0
	Per diem included		
Intrm	2 Trips Cordova/Anchorage @ \$500/trip	\$1.0	\$0.0
	2 RT Anchorage/Juneau @ \$600/trip	\$1.2	\$0.0
	Per diem included		
	6 RT Cordova/Anchorage @ \$500/trip	\$0.0	\$3.1
	4 RT Anchorage/Juneau @ \$500/trip	\$0.0	\$2.4
	Per diem included		
	2 Trips Cordova/Sitka (L Port Walter) @ .9 k -includes per diem		\$1.8
Travel Total		\$7.1	\$7.3
Contractual:			
Intrm	Fall 1993 egg sampling and embryo montoring at AFK		
	Vessel charter	\$25.4	\$0.0
	Air charter	\$3.0	\$0.0
	Vehicle Rental	\$0.6	
	Equipment Rental	\$4.0	
	Facility Lease	\$3.5	
Rem.	Spring 1994 pre-emergent fry survey and AFK embryo collection		\$15.0
	Vessel charter		\$3.6
	Air Charter		\$1.1
	Vehicle Rental		\$1.9
	Equipment Repair		\$1.2
	Facility Lease		\$21.5
	Air Charter for AFK Incubation Experiment		\$15.0
	Genetic Analysis Contract		
Contractual Total		\$36.5	\$59.3

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Project Number: 94191
Project Title: Oil Related Egg & Alevin Mortalities
Sub-Project:
Agency: AK Dept. of Fish & Game

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EXXON VALDEZ TRUSTEE COUNCIL
 1994 Federal Fiscal Year Project Budget
 October 1, 1993 - September 30, 1994

Commodities:		Reprt/Intr	Remaining
Reprt	Laboratory supplies, computer software and upgrades	\$9.5	
Intrm	Field supplies (glassware, water and air pumps, gasoline)	\$4.0	
	Remaining Laboratory Supplies		\$9.5
	Field Supplies		\$4.0
	Fish Culture Suplies		\$4.0
Commodities Total		\$13.5	\$17.5
Equipment:			
Intrm	Microscope objectives for epifluorescent examination of chromosomes	\$2.1	\$0.0
Equipment Total		\$2.1	\$0.0

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Project Number: 94191
 Project Title: Oil Related Egg & Alevin Mortalities
 Sub-Project:
 Agency: AK Dept. of Fish & Game

FORM 3B
SUB-
PROJECT
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EXXON VALDEZ STEE COUNCIL
 1994 Federal Fiscal Year Project Budget
 October 1, 1993 - September 30, 1994

Project Description: Oil Related Egg & Alevin Mortalities - The NOAA/NMFS portion of the egg mortality study provides laboratory verification that field results observed for eggs in 1989, 1990, and 1991 are consistent with immediate lethal effects as well as persistent genetic damage caused by oil deposited in intertidal pink spawning habitat.

Budget Category:	1993 Project No. 93003 Authorized FFY 9	'93 Report/ '94 Interim* FFY 94	Remaining Cost** FFY 94	Total FFY 94	FFY 95	Comment	
						Interim Portion	Report Only Portion
Personnel	\$128.4	\$124.7	\$89.0	\$213.7	\$215.0	\$9.2	\$115.5
Travel	\$11.0	\$5.2	\$16.2	\$21.4	\$20.0	\$2.0	\$3.1
Contractual	\$116.0	\$0.0	\$50.0	\$50.0	\$25.0	\$0.0	\$0.0
Commodities	\$56.0	\$12.7	\$25.8	\$38.5	\$40.0	\$5.7	\$7.0
Equipment	\$19.0	\$0.0	\$15.0	\$15.0	\$5.0	\$0.0	\$0.0
Capital Outlay	\$0.0	\$0.0	\$0.0	\$0.0	\$0.0	\$0.0	\$0.0
Subtotal	\$330.4	\$142.6	\$196.0	\$338.6	\$305.0	\$16.9	\$125.6
General Administration	\$27.4	\$18.7	\$16.9	\$35.6	\$34.0	\$1.4	\$17.3
Project Total	\$357.8	\$161.3	\$212.9	\$374.2	\$339.0	\$18.3	\$142.9
Full-time Equivalents (FTE)	2.8	2.6	1.8	4.4	4.4		
Dollar amounts are shown in thousands of dollars.							
Budget Year Proposed Personnel:		Reprt/Intm Months	Reprt/Intm Cost	Remaining Months	Remaining Cost		
Position Description							
Reprt	Program Manager GS-1 (Int Mo = 0)	0.9	\$4.4	0.6	\$3.1		
&	Project Leader GS-11 (Int Mo = 1)	7.0	\$35.9	5.0	\$25.6		
Intm	Chemist GS-13 (Int Mo = 0)	1.8	\$12.8	1.2	\$9.1		
	Chemist GS-9 (Int Mo = 0)	7.0	\$29.7	5.0	\$21.3		
	Chemist GS-5 (Int Mo = 0)	7.0	\$18.4	5.0	\$13.1		
	Fishery Biologist GS-7 (Int Mo = 1)	7.0	\$23.5	5.0	\$16.8		
Personnel Total		30.7	\$124.7	21.8	\$89.0		
						NEPA Cost:	\$0.0
						*Oct 1, 1993 - Jan 31, 1994	
						**Feb 1, 1994 - Sep 30, 1994	

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Project Number: 94191
 Project Title: Oil Related Egg & Alevin Mortalities
 Sub-Project:
 Agency: National Oceanic & Atmospheric Admin.

FORM 3A
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EXXON VALDEZ TRUSTEE COUNCIL
1994 Federal Fiscal Year Project Budget
October 1, 1993 - September 30, 1994

Travel:		Reprt/Intr	Remaining
Reprt	Travel to field station at Little Port Walter (10 trips x 6 staff)		
&	(7 trips - air fare \$600/trip + 7 days per diem @ \$3/day)	\$4.3	\$0.0
Intrm	(16 trips - air fare \$600/trip + 7 days per diem @ \$3/day)	\$0.0	\$9.9
	Travel between Juneau, Anchorage, and Cordova for inter- and intra-agency discussions among PI's and peer rev	\$0.9	\$6.3
	(8 trips - air fare \$450/trip + 2 days per diem @ \$225/day)		
Travel Total		\$5.2	\$16.2
Contractual:			
	Histopathological examination of emergent fry from 1993 brood to look for tissue abnormalities (cost based on previous contracts)	\$0.0	\$15.0
	Immunofluorescence evaluation of Mixed Function Oxidase induction in emergent fry from 1992 and 1993 broods to indicate exposure to hydrocarbons (cost based on previous contracts)	\$0.0	\$35.0
Contractual Total		\$0.0	\$50.0

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Project Number: 94191
Project Title: Oil Related Egg & Alevin Mortalities
Sub-Project:
Agency: National Oceanic & Atmospheric Admin.

FORM 3B
SUB-
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DETAIL

EXXON VALDEZ STEE COUNCIL
 1994 Federal Fiscal Year Project Budget
 October 1, 1993 - September 30, 1994

Commodities:		Reprt/Intr	Remaining
Reprt	Solvents for hydrocarbon analyses	\$3.5	\$3.0
	Chemistry lab supplies for hydrocarbon analyses	\$3.5	\$5.0
Intrm	Fish food, field supplies	\$3.3	\$3.0
	Groceries for field staff (1 person x 365 days x \$10/day) + (6 persons x 60 days x \$10/day)	\$2.4	\$4.8
	Culture supplies	\$0.0	\$3.0
	Office supplies, software upgrades	\$0.0	\$7.0
Commodities Total		\$12.7	\$25.8
Equipment:			
	PIT tags for identifying individual fish by treatment group (3,750 tags @ \$4/tag)	\$0.0	\$15.0
Equipment Total		\$0.0	\$15.0

07/14/93

1994

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Project Number: 94191
 Project Title: Oil Related Egg & Alevin Mortalities
 Sub-Project:
 Agency: National Oceanic & Atmospheric Admin.

FORM 3B
 SUB-
 PROJECT
 DETAIL

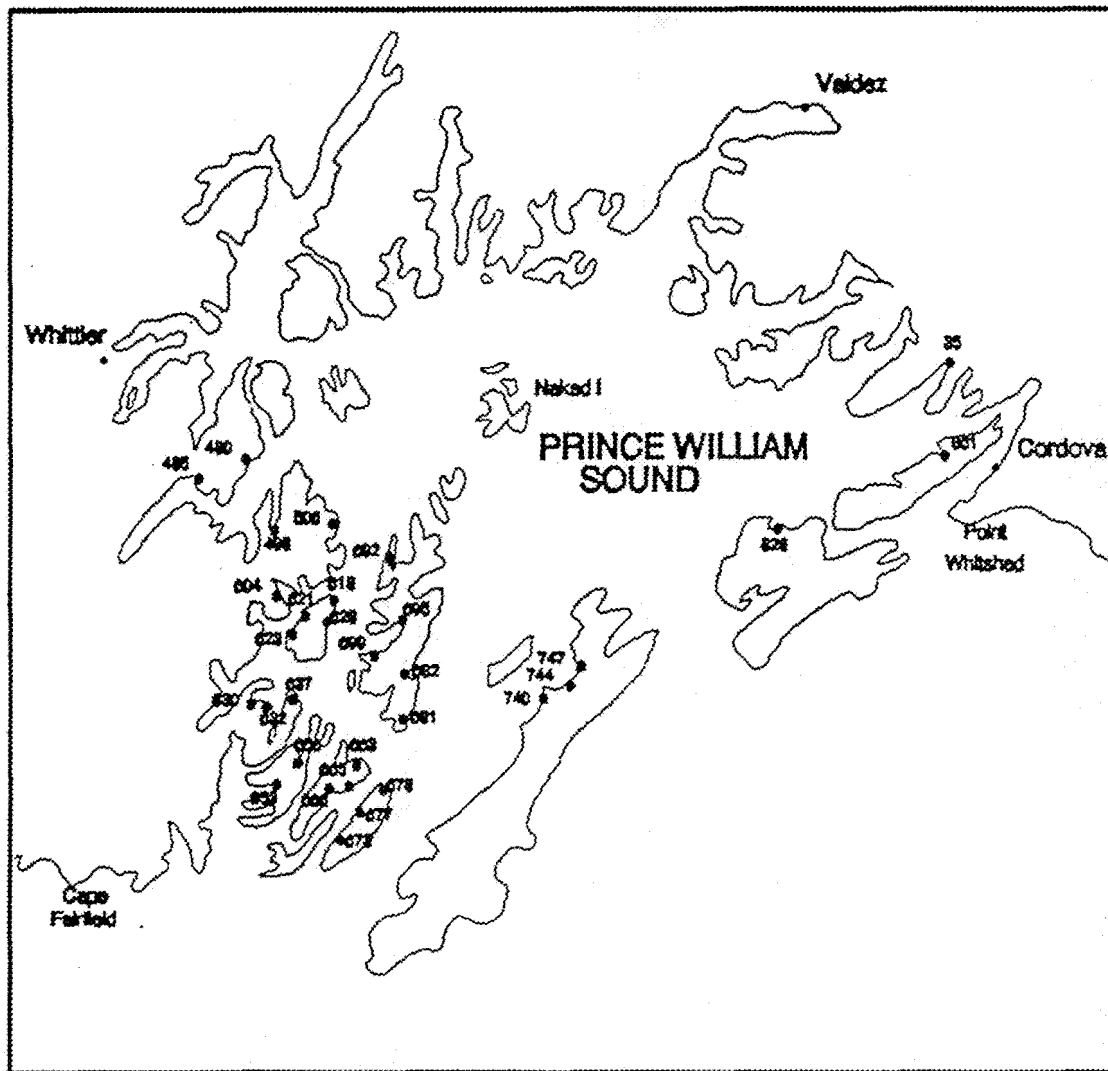


Figure 1. Location of streams to be sampled for egg deposition.

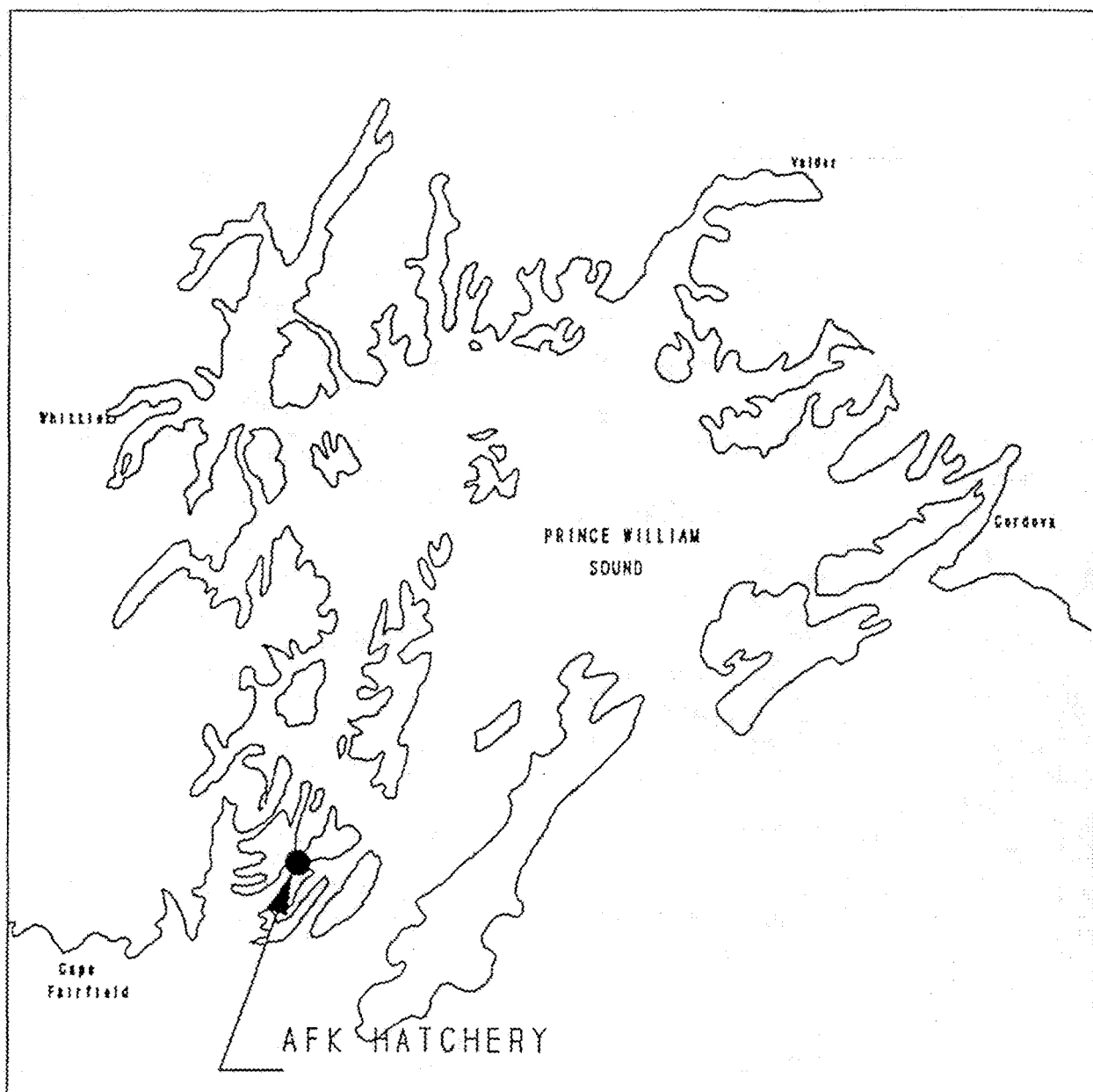


Figure 3. Location of the Prince William Sound Aquaculture, Armin F. Koernig (AFK) Hatchery where eggs will be incubated for Component B of this project.

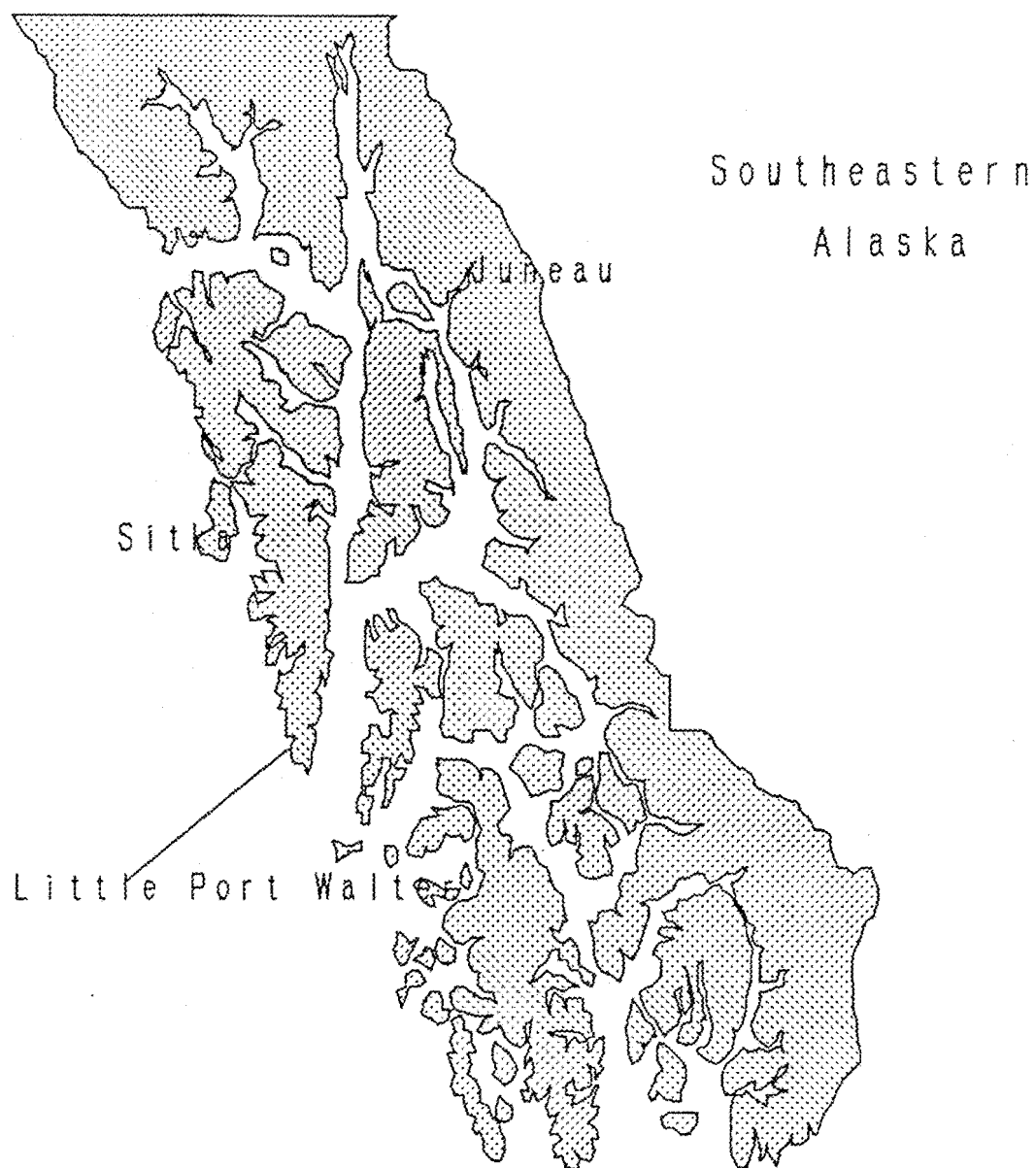



Figure 4. Location of the National Marine Fisheries Service, Little Port Walter facility in southeastern Alaska where Component C Studies will be performed.

DETECTION OF DELETERIOUS MUTATIONS IN PINK SALMON
THROUGH HAPLOID ANDROGENESIS

PROPOSAL TO:

ALASKA DEPARTMENT OF FISH AND GAME
Attn: James Seeb, Principal Geneticist
333 Raspberry Road
Anchorage, Alaska 99518
Phone: (907)-267-2311
Fax: (907)-349-2231

SUBMITTED BY:


Gary Thorgaard, Principal Investigator
Department of Zoology
Washington State University
Pullman, Washington 99164-4236
Phone: (509)-335-7438
Fax: (509)-335-3517

DATE: February 25, 1994

DURATION OF PROJECT: 5/1/94-12/31/94

TOTAL FUNDS REQUESTED: \$15,000
(\$12,500 Direct costs)
(\$2,500 Indirect Costs*)
*a maximum indirect costs rate of
20% of direct costs is allowed
by the Alaska Department of Fish
and Game.

PROJECT SUMMARY:

We will use induced androgenesis to estimate the level of mutations carried by pink salmon in Prince William Sound, Alaska. Androgenesis is a treatment in which eggs are treated with radiation before fertilization, followed by fertilization with normal sperm. The relative survival of androgenetic haploids may be a sensitive measure of the number of harmful mutations carried by sperm from a given male. This study is being carried out to address concerns that some populations may contain high levels of mutations resulting from exposure to mutagenic chemicals.

DETECTION OF DELETERIOUS MUTATIONS IN PINK SALMON THROUGH HAPLOID ANDROGENESIS

We will use induced androgenesis to estimate the level of mutations carried by pink salmon in Prince William Sound, Alaska. This study is being carried out to address concerns that some populations may contain high levels of mutations resulting from exposure to mutagenic chemicals.

Background Information

Androgenesis is a treatment in which eggs are treated with radiation before fertilization, followed by 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. We have studied androgenesis in rainbow trout in our laboratory for a number of years (1-5), primarily as a means to produce clonal lines for research purposes. Such lines can be produced by raising a diploid androgenetic individual to sexual maturity and then producing identical offspring from that individual by another cycle of androgenesis if the androgenetic individual is a male, and by gynogenesis if the androgenetic individual is a female.

The relative survival of androgenetic haploids may be a sensitive measure of the number of harmful mutations carried by sperm from a given male. We prefer to use androgenetic haploids rather than androgenetic diploids because they show better early survival, not having received heat or pressure treatments. Both recessive and dominant mutations should kill haploid embryos, while only dominant mutations will kill normal embryos with one chromosome set from each parent. Recessive mutations are likely to be much more common than dominant ones, hence the haploid assay should be much more sensitive. The survival of androgenetic haploid salamanders has been shown to drop dramatically after male salamanders were exposed to mutagenic chemicals (6). Studies in zebrafish also suggest that harmful genes reduce the survival of haploid individuals (7).

We are currently carrying out experiments in rainbow trout to examine the sensitivity of the androgenetic haploid assay. These preliminary studies are investigating the effect of UV irradiation of sperm on the survival of androgenetic haploids. These studies are comparing the proportions of developing embryos that begin cell division (about 12 hours after fertilization) with the proportion that develop to the eyed stage (about three weeks at 10°C). We are finding that fewer cleaving embryos develop to the eyed stage when the sperm has been exposed to UV light and the effect is dramatically more apparent in haploid androgenetic embryos than in diploid embryos.

Proposed research with pink salmon

Similar studies involving pink salmon will compare the viability of androgenetic haploids produced using sperm from males from an oil-exposed stream where mutations are suspected with the viability of haploids produced using sperm from males from a control stream. The specific streams will be identified by the Alaska Department of Fish and Game (ADF&G). The prediction is that androgenetic haploid offspring will die earlier if the sperm is carrying a high number of mutations. An advantage of this approach is that the same eggs will be used to test the sperm from salmon from the oil-exposed and control stream. Since sperm essentially functions only to carry genes and has virtually no other contribution to the embryo, this experiment will allow us to focus on genetic influences; residual environmental effects should have little chance of influencing the results. Depending on the results in this study and the rainbow trout results, the study could be expanded to additional streams in 1995.

The study will be carried out as follows:

1. Eggs and sperm from pink salmon will be collected in Alaska by ADF&G and the studies will be conducted at Washington State University using the Cobalt-60 gamma source at the WSU Nuclear Radiation Center and our hatchery facilities.
2. We will compare the survival of androgenetic haploid individuals produced from 30 males from an oil-exposed stream with the survival of androgenetic haploid individuals produced from 30 males from a control stream. Each trial will be replicated three times. Using 100 eggs per replicate, a total of about 18,000 unfertilized pink salmon eggs will be used in the experiments, with 6,000 each being used at 4-5 day intervals. The first 6,000 will be brought back with sperm collected in late August and two shipments will follow from Alaska. We will consult with a biometrician from ADF&G about specifics of the experimental design, probably during a visit to Anchorage in May or June.
3. Funds will be used for salary and benefits for Paul Wheeler (the Research Tech II who will carry out the experiments), two trips to Alaska (one for planning and one for collecting gametes), hourly help, lab supplies and overhead.
4. We will be free to publish the results of the study. We understand that the results of the study should not be made public until ADF&G has a chance to review them and they are accepted in a peer-reviewed journal.

References

1. Parsons, J. E. and G. H. Thorgaard, 1984. Induced androgenesis in rainbow trout. J. Exp. Zool. 231: 407-412.
2. Parsons, J. E. and G. H. Thorgaard, 1985. Production of androgenetic diploid rainbow trout. J. Hered. 76: 177-181.
3. Scheerer, P. D., G. H. Thorgaard, F. W. Allendorf and K. L. Knudsen, 1986. Androgenetic rainbow trout produced from inbred and outbred sperm sources show similar survival. Aquaculture 57: 289-298.
4. Thorgaard, G.H., P.D. Scheerer, W.K. Hershberger and J.M. Myers, 1990. Androgenetic rainbow trout produced using sperm from tetraploid males show improved survival. Aquaculture 85: 215-221.
5. Scheerer, P.D., G.H. Thorgaard and F.W. Allendorf, 1991. Genetic analysis of androgenetic rainbow trout. J. Exp. Zool. 260: 382-390.
6. Armstrong, J.B. and W.S. Fletcher, 1983. Assessment of mutagenic damage following ethyl methanesulfonate mutagenesis in the axolotl (Ambystoma mexicanum). J. Exp. Zool 226: 333-340.
7. Streisinger, G., C. Walker, N. Dower, D. Knauber and F. Singer, 1981. Production of clones of homozygous diploid zebrafish (Brachydanio rerio). Nature 291: 293-296.

BUDGET

Submitted to: Alaska Department of Fish and Game
Period of Project: 5/1/94-12/31/94

Salaries and Wages

Fac: G.H. Thorgaard, Prof. N/C

Stf: Research Tech II (Paul Wheeler)
4 mos @ 50% (8/16/94-12/15/94) 5,066

Hourly Help (\$7/hr. X 170 hrs.) 1,190

Total Salaries & Wages 6,256

Benefits

PW 1,317
Hourly help 18

Total Benefits 1,335

Total Salaries, Wages and Benefits 7,591

Equipment -

Supplies 2,909

Travel 2,000

(Two trips to Alaska)

Other Costs -

Total Direct Costs 12,500

Indirect Costs 2,500

(@ 20% of direct costs, maximum rate allowed by ADF&G)

TOTAL PROJECT COSTS - 15,000

EXXON VALDEZ TRUSTEE COUNCIL
FY 94 DETAILED PROJECT DESCRIPTION

Project title: Oil Related Egg and Alevin Mortalities

Project ID number: 94191 - Supplemental

Project type: Monitoring and Research

Name of project leaders: Samuel Sharr, Alaska Dept. Fish and Game
Jim Seeb, Alaska Dept. Fish and Game

Lead agency: Alaska Department of Fish and Game

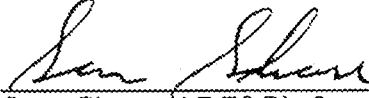
Cooperating agencies: National Marine Fisheries Service
Other Cooperating Parties: Washington State University

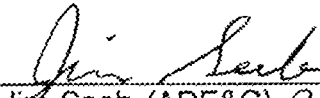
Cost of project/FY 94: \$ 97.7
Cost of project/FY 95: \$128.4
Cost of Project/FY 96 and beyond: \$130.4

Project Start-up/Completion Dates: October 1, 1994 to September 30, 1996


Geographic area of project: Prince William Sound

Project leaders:


Sam Sharr (ADF&G) By Brian Bue 3/18/94
Date


Jim Seeb (ADF&G) By Brian Bue 3/18/94
Date

Agency project managers:


Joe Sullivan (ADF&G) 3/18/94
Date

B. INTRODUCTION

This project was included in the 1993 Work Plan under project 93003. At the time proposals were submitted for the 1994 work plan, we elected to discontinue the work proposed here. This decision was based on our perception that oil damages had diminished to an undetectable level; consequently, we believed this work had very little probability of providing additional information on damages. When preliminary results of the 1993 field season were obtained in December of 1993, we found that this project provided strong evidence for the existence of genetic damage. This project will incorporate this work back into the 1994 work plan.

Summary

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 have 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 (Sharr et al. 1994a, 1994b, and in prep; and Bue et al. in press). The consequences of this putative genetic damage include physiological dysfunction and functional sterilization of affected individuals, reducing the 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.

This study will continue to monitor the recovery of pink salmon embryos and provide an assessment of the role that physical stream characteristics played in the damages observed by Sharr et al. (1994a and 1994b) and Bue et al. (in press) in the field. This will be accomplished by collecting pink salmon gametes from oil contaminated and uncontaminated streams in southwestern Prince William Sound (PWS) and incubating them under identical controlled conditions.

History

Pink salmon eggs and fry incubating in the oiled intertidal spawning areas in Prince William Sound in 1989, 1990, 1991, and 1992 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 gravels 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 pink salmon egg mortalities in the intertidal zones of oiled streams while no difference between oiled and non-oiled streams was detected above mean high tide. Elevated egg mortalities in oiled streams were again detected in the 1990 brood year, but only in the highest intertidal spawning zone (Sharr et al. 1994a, and 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 egg 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 egg mortalities in oiled streams during the fall of 1991 survey. Furthermore, significant differences in egg mortality occurred at all tidal zones, including the area above mean high tide. Clearly, the elevated egg 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 egg mortalities in 1991 may be the result of genetic damage acquired during development after fertilization in 1989. Elevated egg mortalities at all tidal zones in oiled streams were again detected during the fall of 1992 survey (Sharr et al. 1994b and 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 sampling design (Sharr et al. in prep).

The hypothesis of genetic damage is consistent with previous laboratory experiments on the effects of crude oil on early life stages of fish and with other NRDA field observations. 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 eggs and larvae exposed to very low aqueous doses (1 ul oil/l seawater) of oil. Moles et al. (1987) confirmed that pink salmon eggs 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 reproductive fitness of a significant proportion of exposed individuals.

Information gained from this study will provide resource managers insight to the magnitude and persistence of damages sustained by wild pink salmon due to EVOS. Efforts to restore damaged pink salmon populations depend upon the fishery manager's abilities to identify sources of reduced survival and to monitor their persistence. Information on the potential of long term oil exposures to cause genetic damage is needed so spawning escapement goals can be reevaluated and adjusted if necessary. In addition, verification of the genetic

hypothesis would provide the first evidence that reproductive capacity of fish exposed to chronic or acute sources of oil pollution would be compromised.

C. PROJECT DESCRIPTION

This project is an addition to project 94191 - *Oil Related Egg and Alevin Mortalities*. In this study we will collect pink salmon gametes from adult pink salmon returning to oil contaminated and unimpacted streams in western Prince William Sound (PWS). Intra-stream crosses will be made and the resulting embryos incubated under identical controlled conditions to evaluate the effect of physical stream characteristics.

This project will also administer a contract (15k) for a pilot study to examine the usefulness of androgenesis for evaluating genetic damage in pink salmon. Androgenetic individuals are obtained by enucleating eggs with gamma radiation before fertilization. These eggs are then fertilized with normal sperm. If no other treatments are applied, the resulting progeny will be haploid, containing only a single set of chromosomes from the male parent and none from the female. Mortality rates for these haploids are directly related to the presence and number of deleterious mutations (Armstrong and Fletcher, 1983). Advantages of this technique over most 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. Ultimately, haploid androgens will be used to test for the presence of deleterious mutations in the chromosomes of oil impacted and control populations in Prince William Sound as well as oil treated and control populations from the Little Port Walter experiment (component B of Project 94191).

1. Resources and/or Associated Services:

This study will investigate pink salmon in Prince William Sound, Alaska. Pink salmon are a major predator and prey species in the PWS ecosystem and provide transport of nutrients from the marine to terrestrial ecosystem. Pink salmon also support large commercial, sport, and subsistence fisheries which are vital to the economy of the area.

2. Relation to Other Damage Assessment/Restoration Work:

The foundations for this project date back to the original NRDA F/S Study 2 (Injury to Salmon Eggs and Preemergent Fry). NRDA F/S Study 2 was equivalent to the field monitoring portion of Project 94191 (Component A of 94191) and was conducted in 1989, 1990, and 1991. The same project was continued as Restoration Study R60C in 1992. Two additional elements, a controlled oiling experiment (Component B of 94191) and the study addressed in this proposal were added to Restoration Study R60C during the summer of 1992. These additions were designed to assess the genetic damage hypotheses raised through NRDA F/S Study 2. All three components were present in the 1993 project, Restoration Study 93003.

At the time proposals were submitted for the 1994 work plan, we elected to discontinue the work being proposed here. This decision was based on our perception that oil damages had diminished to an undetectable level; consequently, we believed this work had very little probability of providing additional information on damages. When preliminary results of the 1993 field season were obtained in December of 1993, we found that this project provided strong evidence for the existence of genetic damage. It was at that time that we began to work towards putting this study back into the 1994 work plan.

Several past NRDA and present Restoration projects have been and continue to be intimately related to this project. The 1989 and 1990 NRDA F/S Study 4 demonstrated reduced growth and survival for salmon which reared in oiled areas. NRDA F/S Study 1 in 1989, 1990, and 1991 and subsequent Restoration Study R60B in 1992, investigated oil damage to adult pink salmon spawning populations and provided valuable improvements in escapement estimation procedures used by fisheries managers to monitor and protect injured wild pink salmon populations. NRDA F/S Study 3 in 1989, 1990, and 1991 and subsequent Restoration studies R60A in 1992 and 93185 in 1993 provided hatchery and wild catch contribution estimates. This information was used by fisheries managers to reduce fisheries exploitation rates on injured wild pink salmon and also provided survival estimates for groups of fish examined by NRDA Study 4. The 1989, 1990, and 1991 NRDA F/S Study 28 and a subsequent Restoration study in 1992, incorporated data from all the previous studies into life history and run reconstruction models. These models were used to extrapolate losses in adult pink salmon production from injuries observed in earlier life history stages.

3. Objectives:

1. Determine whether the increased pink salmon egg mortalities observed in oiled streams can be attributed to the physical characteristics of the study streams.

4. Methods:

1. Experimental Design

The experiment will assess the effects of the physical characteristics of the study streams upon the observed results. This will be accomplished by collecting pink salmon gametes from oiled and non-oiled streams and rearing the resulting embryos in a controlled laboratory environment.

This experiment will provide information to help determine whether the results observed in NRDA Study FS2 can be attributed solely to the physical characteristics of the study streams. In this experiment we will collect gametes from 8 oiled and 8 non-oiled streams from southwestern PWS, make intra-stream crosses, and incubate the resulting embryos in a controlled laboratory environment. Egg mortality will be compared between the oiled and uncontaminated streams. If no difference is observed in this experiment and a significant difference in egg mortality is detected between oiled and non-oiled streams during the recovery monitoring portion of this study during the fall of 1993 egg sampling, it can be stated that the physical characteristics of the study streams played a role in the results of the previous egg mortality studies.

Gamete collection and fertilization procedures will occur over a four day period to obtain data from 8 oiled and 8 non-oiled streams. Gametes from 30 male and 30 female pink salmon will be collected from 2 oiled and 2 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 should insure that all possible crosses ($30 \times 30 = 900$) 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 (Heath Incubator).

Incubating embryos will be periodically screened for dead eggs and hatching success. Samples of sperm from each male used to build the embryo pools will be cryopreserved for future analysis if required. The experiment will be terminated prior to swimup at which time all larvae will be killed.

2. Data Analysis

The data will be analyzed as a fixed-effects generalized randomized block design:

$$Y_{ijk} = \mu + B_i + O_j + \epsilon_{ijk} \quad (1)$$

where Y_{ijk} is egg mortality for sample day i , oil contamination level j , and stream k ; μ 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 ϵ_{ijk} is random error. The relative power of the test was estimated. The sample size was considered sufficient to detect a difference of less than 1.5 standard deviations at $\alpha = 0.05$ and 95% power (Neter et al. 1990). A test with high power is needed to protect against arriving at the conclusion that all observed damages could be attributed to the physical characteristics of the streams when in actuality significant damages due to oil were present.

The assumption of constant error terms will be tested using the F_{\max} -test (Sokal and Rohlf 1969) 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.

5. Location:

This study will collect gametes from streams in southwestern Prince William Sound and incubate the resulting embryos at the Armin F. Koernig hatchery (Figure 1).

6. Technical Support:

A biometrician will ensure the study design will provide a reasonable chance of reaching a defensible conclusion.

7. Contracts:

We propose to have a \$15,000 project *Detection of Deleterious Mutations in Pink Salmon Through Haploid Androgenesis* conducted by Dr. Gary Thorgaard, Washington State University (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. Dr. Thorgaard's proposal is attached.

D. SCHEDULES

Dates	Activity
30 Oct 1993 - 30 Jan 1994	Analysis of 1993 data and completion of first draft of 93003 report for laboratory evaluation
April 1994	Initiate Androgenesis Contract
1 Aug - 15 Aug 1994	Preparation for 1994 AFK Incubation Experiment
15 Aug - 30 Aug 1994	Collect Gametes and make crosses from 16 PWS streams; begin incubation of gametes at AFK.
30 Aug - 15 Nov 1994	Monitor incubators and collect data
15 December 1994	Androgenesis Contract Report Due to ADF&G
15 Nov 1994 - 30 Jan 1995	Analyze data and prepare first draft of 94191 report

E. EXISTING AGENCY PROGRAM

This project will benefit from both ADF&G's commercial fisheries management and genetics programs. Both groups provide supporting information for the successful completion of the project.

F. ENVIRONMENTAL COMPLIANCE/PERMIT/COORDINATION STATUS

Transport of wild gametes to the Prince William Sound Aquaculture Corporation (PWSAC) hatchery on Evans Island, PWS will require an Alaska Department of Fish and Game (ADF&G) Fish Transport Permit for each stock and a permit Alteration may be required to rear and incubate the wild eggs at the hatchery.

G. PERFORMANCE MONITORING

Principal investigator Sharr (Fisheries Biologist III) will help design the experiment, supervise incubator setup, and coordinate and supervise field logistics. Principal investigator Seeb (Principal Geneticist) will help design and provide genetics oversight as well as administer the proposed androgenesis contract. Seeb's assistant, Gary Miller (Fisheries Biologist II) will provide fish culture oversight and will supervise the technicians responsible for collecting the data. Consulting biometrician Bue (Biometrician II) will conduct the experimental design and provide statistical oversight for the project. Sharr, Seeb, and Bue will cooperate in the data analysis and writing of the project report.

The methodologies for this project have been approved by the Chief Scientist and his staff in past proposals. Past work has shown the methods to be appropriate and efficient. The principal geneticist and his staff have extensive laboratory fish culture experience and will be present at all times during the rearing experiment at AFK Hatchery.

H. COORDINATION OF INTEGRATED RESEARCH EFFORT

Data collection for this project occurs over a very compressed period of time and is very stream specific; hence, this study does not blend well logistically with most of the other pink salmon projects. However, all of the streams used for brood stock in this study are also of interest to the Pink Salmon Genetics Project (94189) and carcasses from the egg takes can be used for genetic samples from these streams. The study is housed at the Prince William Sound Aquaculture Corporation (PWSAC) Armin F. Koerning (AFK) hatchery and will take advantage of the same incubation facilities as the PWSAC experimental release project included in the SEA project.

Final edited data 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.

I. PUBLIC PROCESS

Many of the field procedures used in the field monitoring portion (Component A of 94191) of this project have been employed as part of the data collection activities for preemergent fry indices used in PWS pink salmon forecasts for more than 30 years. The procedures have been presented and reviewed at a multitude of workshops and scientific meetings, are widely understood by the fishing industry, and have undergone peer review through the NRDA process. Field monitoring methodologies were presented at the 1991 Pink and Chum Workshop in Parksville, British Columbia, Canada. Field monitoring results from 1989, 1990, 1991, and 1992 were presented at the 1993 meeting of the Alaska Chapter of The American Fisheries Society in Valdez, Alaska, the 1993 Oil Spill Symposium in Anchorage, Alaska, and the 1993 Pink and Chum Workshop in Juneau, Alaska. Abbreviated operational plans for 1989 through 1994 egg and alevin mortality studies have been published annually in EVOS Trustee Council work plans which incorporate public comment.

J. PERSONNEL QUALIFICATIONS

Fisheries Biologist III - Samuel Sharr

Mr. Sharr received a Bachelor of Science degree in biology from the University of Washington in 1968. He has been a research biologist for ADF&G since 1979 and has worked on PWS salmon and herring since 1981. He assumed his present position as the ADF&G, Division of Commercial Fisheries, Biologist III, PWS Area Finfish Research Project Leader in 1986. In this capacity, Mr. Sharr oversees all the salmon and herring research conducted by the Division of Commercial Fisheries in PWS. His involvement with the PWS salmon escapement aerial survey program dates from the early 1980's. Mr. Sharr has supervised a total re-edit of the historic aerial and ground survey data and designed a new RBASE data base for inseason escapement analyses. Mr. Sharr wrote the original operational plans for NRDA F/S Studies 1,2 and, 3 and has been the Principal Investigator for those projects since their inception.

Principal Geneticist - James E. Seeb

Jim Seeb earned a B.S. in Biology (1974) from the University of Puget Sound, an M.S. in Fisheries (1982) and a Ph.D. in Fisheries (1987) from the University of Washington. Jim has worked as a Fish Biologist for the Washington Department of Fisheries (1978-1980) and Pacific Fisheries Research (1980-1982), as a Graduate Research Assistant at the University of Washington (1982-1986), a Research Assistant Professor at the University of Idaho (1987-1988), and as an Assistant Professor at Southern Illinois University (1988-1990). Presently, Jim is the Principal Geneticist for FRED Division of the Alaska Department of Fish and Game and has overall responsibility for fisheries genetic issues throughout Alaska. Dr. Seeb has published extensively in the Fisheries and Genetics Literature. He has worked with many fish species on numerous genetic topics including but not limited to genetic marking and its use to assess stock dynamics and management programs, genetic variation and postglacial dispersal of populations, the use of genetic structure in the enforcement of fishing regulations, and the measurement of DNA content using flow cytometry.

Biometrician II - Brian G. Bue

Brian Bue has a Bachelor of Science in Biology and a Bachelor of Science in Fisheries from the University of Alaska, Fairbanks. He also possesses a Masters degree in Fisheries with an emphasis on quantitative studies from the University of Alaska, Fairbanks. Brian has worked with the ADF&G from 1974 through present in many capacities. He has worked as a consulting biometrician on oil spill damage assessment projects since the first days of the *Exxon Valdez* spill.

Fisheries Biologist II - Gary Miller

Gary Miller is the flow cytometry specialist for the Alaska Department of Fish and Game Genetics Laboratory in Anchorage. Gary has a Bachelor of Science in Fisheries Biology from the University of Washington, a M.S. in Zoology from Southern Illinois University - Carbondale, and is currently pursuing his Ph.D. from the University of Washington. He has worked periodically for the Alaska Department of Fish and Game since 1981. He has a strong background in genetics and developmental biology and has conducted research and co-authored projects in hybridization, polyploid induction, allozyme expression, and growth performance of triploid salmonids and other fishes. He has extensive laboratory experience with techniques including flow cytometry, protein starch gel electrophoresis, protein and molecular marker analysis, and fluorescent antibody testing of pathogens.

K. BUDGET

(see attached)

L. LITERATURE CITED

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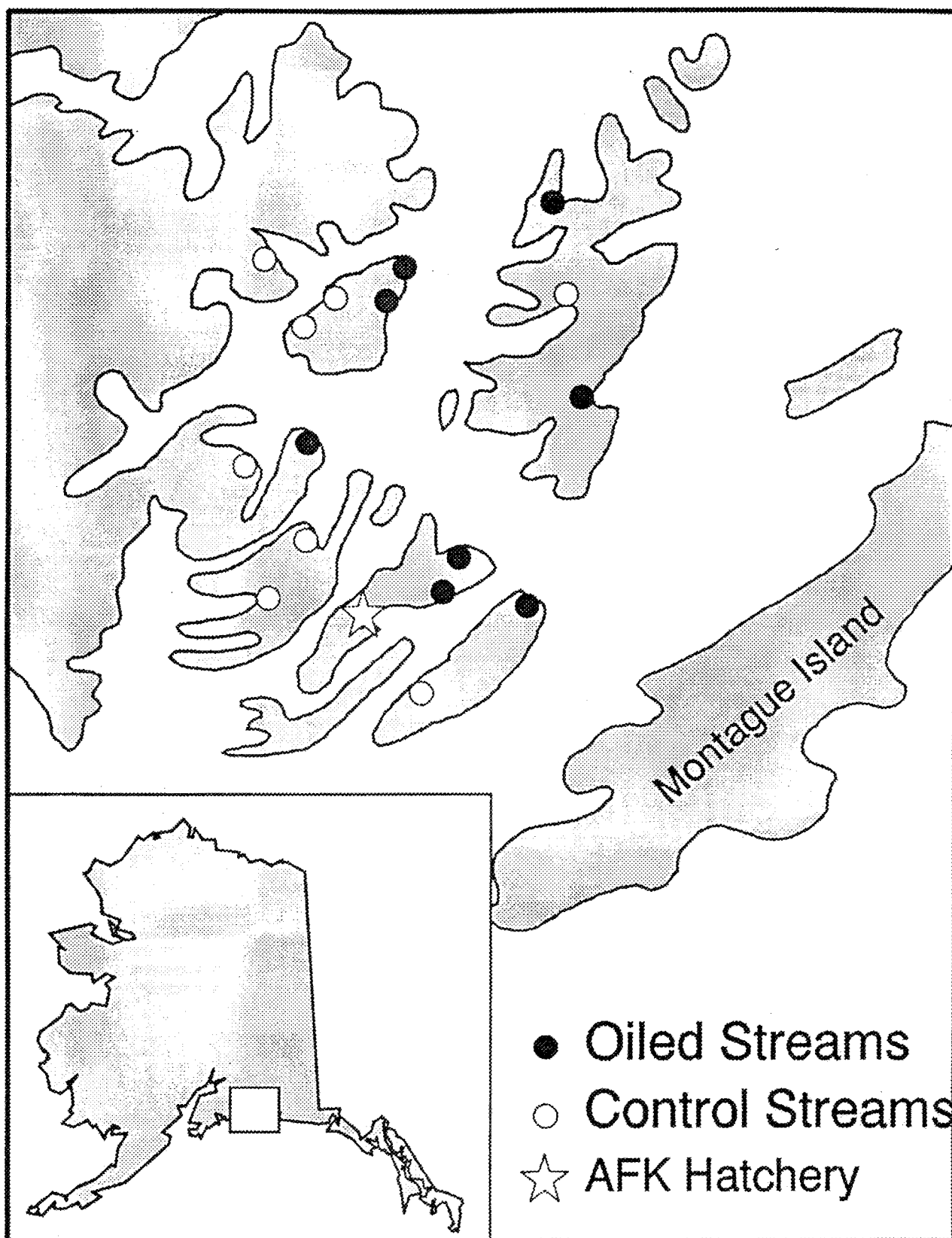


Figure 1. Stream and Hatchery locations for controlled incubation experiment.

EXXON VALDEZ TRUSTEE COUNCIL
1994 Federal Fiscal Year Project Budget
October 1, 1993 - September 30, 1994

Project Description: Oil Related Egg & Alevin Mortalities (Supplemental) - This project will evaluate whether the differences in pink salmon embryo mortality observed in the field under Component A of Project 94191 can be attributed to differences in physical stream makeup or to genetic differences. This experiment will examine the possibility of genetic injury as an explanation for chronic injury and assess the likely time frame for natural recovery.

Budget Category:	1993 Project No. 93003 Authorized FFY 93	'93 Report/ '94 Interim* FFY 94	Remaining Cost** FFY 94	Total FFY 94	FFY 95	Comment
Personnel		\$0.0	\$44.7	\$44.7	\$65.0	
Travel		\$0.0	\$2.0	\$2.0	\$3.5	
Contractual		\$0.0	\$37.7	\$37.7	\$42.7	
Commodities		\$0.0	\$4.0	\$4.0	\$4.5	
Equipment		\$0.0	\$0.0	\$0.0	\$0.0	
Capital Outlay		\$0.0	\$0.0	\$0.0	\$0.0	
Subtotal		\$0.0	\$88.4	\$88.4	\$115.7	
General Administration		\$0.0	\$9.3	\$9.3	\$12.7	
Project Total		\$0.0	\$97.7	\$97.7	\$128.4	
Full-time Equivalents (FTE)		0.0	0.7	0.7	1.1	
Dollar amounts are shown in thousands of dollars.						
Budget Year Proposed Personnel:		Reprt/Intrm Months	Reprt/Intrm Cost	Remaining Months	Remaining Cost	
Position Description						
Fisheries Biologist III				1.5	\$9.9	
Fisheries Biologist II				2.7	\$14.6	
Fish and Wildlife Technician III				2.0	\$6.7	
Fish and Wildlife Technician II				0.6	\$4.3	
Biometrician II				1.5	\$9.2	
Personnel Total		0.0	\$0.0	8.3	\$44.7	
					NEPA Cost:	\$0.0
					*Oct 1, 1993 - Jan 31, 1994	
					**Feb 1, 1994 - Sep 30, 1994	

07/14/93

1994

Project Number: 94191 - Supplemental
Project Title: Oil Related Egg & Alevin Mortalities
Sub-Project:
Agency: AK Dept. of Fish & Game

FORM 3A
SUB-
PROJECT
DETAIL

EXXON VALDEZ TRUSTEE COUNCIL
 1994 Federal Fiscal Year Project Budget
 October 1, 1993 - September 30, 1994

Travel:	Reprt/Intrm	Remaining
4 Trips Cordova/Anchorage @ \$500/trip Per diem included		\$2.0
Travel Total	\$0.0	\$2.0
Contractual:		
Facility Lease		\$1.2
Air Charter		\$21.5
Genetic Analysis Contract		\$15.0
Contractual Total	\$0.0	\$37.7

07/14/93

1994

Project Number: 94191 - Supplemental
 Project Title: Oil Related Egg & Alevin Mortalities
 Sub-Project:
 Agency: AK Dept. of Fish & Game

FORM 3B
 SUB-
 PROJECT
 DETAIL

EXXON VALDEZ TRUSTEE COUNCIL
 1994 Federal Fiscal Year Project Budget
 October 1, 1993 - September 30, 1994

Commodities:		Reprt/Intrm	Remaining
Fish Culture Supplies			\$4.0
Commodities Total		\$0.0	\$4.0
Equipment:			
Equipment Total		\$0.0	\$0.0

07/14/93

1994

Project Number: 94191 - Supplemental
 Project Title: Oil Related Egg & Alevin Mortalities
 Sub-Project:
 Agency: AK Dept. of Fish & Game

FORM 3B
 SUB-
 PROJECT
 DETAIL

**DETAILED PROJECT DESCRIPTION
IS NOT APPLICABLE FOR THIS PROJECT
BRIEF PROJECT DESCRIPTION FOLLOWS**

EXXON VALDEZ OIL SPILL PROJECT DESCRIPTION

Title: Alaska Marine Research Institute

Project Number: 94199

Lead Agency: ADF&G

Cooperating Agencies: NOAA and DOI-FWS

Cost of Project, FY94: To be determined

Cost of Project, FY95: To be determine

Cost of Project, FY96: To be determine

Project Startup Date: February 1994 **Duration:** 4 years

Geographic Area: Spill Area

INTRODUCTION

The goal of this project is to provide the planned Alaska Marine Research Institute with the scientific equipment and materials necessary to meet monitoring, restoration, rehabilitation and research objectives that promote long-term recovery and health of the marine ecosystem injured by the *Exxon Valdez* oil spill. This Institute will be jointly planned by the University of Alaska, National Oceanic and Atmospheric Administration, Alaska Department of Fish and Game, City of Seward and the United States Fish and Wildlife Service. Information learned through the research Institute would be useful not only in the oil spill area of Alaska but throughout northern latitudes.

This marine research institution will provide a unified northern marine ecosystem program of marine research and monitoring. The Institute will provide a needed research facility for restoration work related to the spill. Operational support for the facility will come from project grants secured by staff scientists, from public donations and from other sources.

In 1993, the Alaska State Legislature appropriated from the *Exxon Valdez* criminal restitution fund \$12.5 million to the Alaska Department of Administration for payment as a grant under AS 37.05.315 to the City of Seward "for development of ... a center for education and research related to the natural resources injured by the *Exxon Valdez* oil spill" (Sec. 2, Ch. 79, SLA 1993). This money will be used to initiate construction. A master business plan is being prepared for the Alaska Marine Research Institute in consultation with the University of Alaska. The Trustee Council is being requested to provide funding for the acquisition and installation of laboratory equipment and materials for research.

PROJECT DESCRIPTION

The Alaska Marine Research Institute is planned as an on-going scientific research institute which will, as a central part of its mission, enhance and provide facilities and personnel, necessary to meet monitoring, restoration rehabilitation and research objectives that promote long-term recovery and health of the marine ecosystem injured by the oil spill. It would be located on waterfront property previously donated by the City of Seward. A preliminary design of the center was prepared by Cambridge Seven Associates. The design anticipates that the Alaska Marine Research Institute will operate in cooperation with the University of Alaska-Fairbanks' Seward Marine Center. Operated by the Institute of Marine Sciences, the Seward Marine Center has an international reputation as a unique subarctic research institution devoted to fish, invertebrates, and medical research.

The design for the Institute will include marine mammal pools of varying sizes to hold different species of marine mammals and pools to accommodate diving and wading birds. A 5,000ft² building will provide wet and dry labs that would enable researchers to bring birds and mammals into a controlled environment. Office space and state-of-the-art data processing facilities and equipment will also be available for researchers and graduate students. The design will also include a variety of tanks, pools and cages of varying sizes that will be available to house marine mammals, seabirds, fish and invertebrates. A 5,000ft² hospital containing a medical treatment center, small clinical laboratory, and intensive care pens for pinnipeds, otters, and seabirds will be part of the rehabilitation area.

It is expected that many of the animals housed in these facilities will be of great interest to the public. Though the Trustee Council is not being asked to fund a visitor center or public aquarium, the facilities could be configured to accommodate this aspect also if funding can be obtained from other sources.

A. Resources and/or Associated Services

The Alaska Marine Research Institute would aid recovery of marine mammals, seabirds and other marine life injured by the *Exxon Valdez* oil spill through facilitating research, monitoring, restoration and rehabilitation of these species. The recovery of harbor seals and marbled murrelets is particularly uncertain because their populations were in decline before the oil spill occurred.

In the course of pursuing the restoration of the spill-impacted ecosystem, the Institute will provide ancillary benefits not specifically targeted by restoration projects. The presence of the Institute will promote interest in and knowledge about the marine and coastal ecosystem, especially its resources affected by the spill, and create and nurture expertise needed for understanding Alaska's marine environment.

Project Description

B. Objectives

The Alaska Marine Research Institute is planned as an on-going scientific research institute which will, as a central part of its mission, enhance and provide facilities and personnel necessary to meet monitoring, restoration rehabilitation and research objectives that promote long-term recovery and health of the marine ecosystem injured by the oil spill. The portion of this project that the Trustees are being asked to fund and which assists in meeting this goal is the acquisition and installation of the state-of-the-art scientific equipment and materials needed to make the Institute a world class research and restoration facility. It is the intent of this proposal being presented to the Trustee Council to request funding for only that portion of the Alaska Marine Research Institute project which is consistent with the spending guidelines for the civil settlement monies set forth in the Memorandum of Agreement and Consent Decree filed with the United States District Court for the District of Alaska, United States vs. State of Alaska, A91-081. Acquisition and installation of scientific equipment and materials used for the conduct of restoration, research and monitoring projects appears to be consistent with that decree.

C. Methods

The Alaska Marine Research Institute will be developed in four phases.

- Phase I: Project identity, master plan, and economic evaluation (in progress)
- Phase II: Pre-construction, scoping and design (March 1994)
- Phase III: Site development and detailed design (Summer 1994)
- Phase IV: Construction and acquisition and installation of research equipment and materials (Summer 1994 - Spring 1997)

Funding will be provided by a number of sources including the criminal settlement fund, joint trust funds, and private contributions. The total cost of the project has not yet been determined. Funding for Phase II, Phase III and the construction portion of Phase IV has been committed through the \$12.5 million legislative appropriation and private contributions. Joint Trust Funds would finance the acquisition and installation of equipment and materials part of Phase IV.

D. Location

The Alaska Marine Research Institute will be located in Seward. Its services will be available to the entire spill area and the North Pacific region.

E. Technical Support

None required.

F. Contracts

The State is expected to award a grant to the City of Seward for the use of these funds to construct the research and restoration facility. No other state contracts are anticipated.

SCHEDULES

The goal of the Institute's sponsors is to open the doors of the Alaska Marine Research Institute in Spring 1997. In FY94, restoration funds would be used primarily for identification, scoping and design of lab facilities, equipment and materials. This effort would begin in February 1994 and conclude in September 1994. In FY95, FY96 and FY97, facility construction, and acquisition and installation of research equipment and materials would occur. Staffing and start-up would occur as facility completion warrants and as funding becomes available.

EXISTING AGENCY PROGRAM

The Alaska Marine Research Institute would be located adjacent to and cooperate with the Institute of Marine Science's Seward Marine Center. Research, monitoring, rehabilitation and restoration efforts will operate with the advice and cooperation of the National Oceanic and Atmospheric Administration, the Alaska Department of Fish and Game and U.S. Fish and Wildlife Service.

COMPLIANCE/PERMIT/COORDINATION STATUS

The master plan, which will be completed about February 1994, will identify all permits required. The project is expected to require a Section 404 permit from the U.S. Army Corps of Engineers and may require authorization from the Environmental Protection Agency (NPDES permit) for water discharge, depending on the type of water system selected for the facility. Because a decision to fund this project is considered a major federal action, it will require formal documentation of compliance with the National Environmental Policy Act. Depending on the outcome of the environmental assessment, an Environmental Impact Statement may be required. On behalf of the Trustee Council, the National Oceanic and Atmospheric Administration will be the lead agency for compliance with the National Environmental Policy Act.

PERFORMANCE MONITORING

Periodic reports from the project manager and financial audits will be the primary means of monitoring performance.

FY94 BUDGET (\$K)

Budget amounts for the 1994 fiscal year have yet to be determined. NEPA costs are estimated to be \$5K.

94217

**NO DETAILED PROJECT DESCRIPTION REQUIRED --
PROJECT IS A CONTINUATION OF
PREVIOUSLY APPROVED WORK**

BRIEF PROJECT DESCRIPTION FOLLOWS

EXXON VALDEZ OIL SPILL PROJECT DESCRIPTION

Title: Prince William Sound Area Recreation Implementation Plan

Project Number: 94217

Lead Agency: USDA-FS

Cooperating Agency: ADNR

Cost of Project, FY94: \$91.2K

Cost of Project, FY95: \$0.0K

Project Startup Date: October 1993

Duration: 8 months

Geographic Area: Prince William Sound

INTRODUCTION

This project is the continuation and completion of project 93065-Prince William Sound Recreation. The objectives of the project are to develop a list of prioritized recreation restoration projects, identify and describe potential special designations, and identify real or perceived injury to the recreation resource and services in Prince William Sound (PWS). Restoration management goals and objectives for recreation in PWS will also be developed based on injuries identified. This project is currently in the public participation stage with additional public participation and meetings planned through November, 1993.

PROJECT DESCRIPTION

A. Resources and/or Associated Services

This project is focused on restoration of recreation. The most significant part of this project will identify, prioritize, and describe potential recreation restoration projects through public participation. Meetings with interested groups and individuals have taken place with additional meetings scheduled through mid-September. A workshop is planned for November 5 and 6, 1993, which will bring all interested parties together to develop a project list. Evaluation criteria have been developed for use at this meeting to rate and prioritize projects. Additional meetings are being scheduled in the Anchorage area.

In addition to developing projects, potential areas requiring special designation are being identified. A preliminary list of possible designations has been developed. During public meetings, comments about special designations are being gathered. The final project report will include the findings of the special designation research.

A specific statement of injury to recreation in PWS will be developed. This statement will be used as the basis for future recreation restoration activities. Currently, the project has identified existing management goals and objectives from public land management plans. These tend to be broad in scope and not specific to recreation restoration.

B. Objectives

The objectives of this project remain the same as in 1993, with only slight modification in emphasis:

1. Develop recreation resource and service projects for FY 1994 and beyond that can be implemented by the Trustee Council for restoration in PWS. An initial list of projects with complete project descriptions will be provided by November 22, 1993 for inclusion in the FY94 Work Plan. Additional projects will be identified, prioritized, and listed in the final project report.
2. Identify all possible special designations that could be applied to PWS. This would include State, Federal or other yet unknown designations. A preliminary draft of possible special designations will be available September 30, 1993 with the summary of findings included in the final project report.
3. Identify the real or perceived injury to recreation in PWS and develop long-term recreation restoration goals and objectives for the management of PWS. A statement of the injury to recreation will be completed. Goals and objectives will be identified in the project report.

C. Methods

The methods described in the 1993 project description (93065) are being implemented. A mass mailing was sent out in early June to inform people of the project and begin the process of soliciting comment and project ideas. In Addition, people were asked if they would like to fill out the *Exxon Valdez* Restoration Plan Recreation Questionnaire. Returned questionnaires have been added to the data base. Public meetings soliciting ideas from people on possible recreation restoration projects are ongoing with meetings planned through September. Initial work has been done to develop the special designations report and identify recreation restoration management goals and objectives. Information collected at each public meeting is being recorded and will be summarized before the workshop in November and for the project report.

The implementation phase of projects for recreation restoration will not be managed by this project work group. After the November workshop, project descriptions will be prepared for the highest rated projects and submitted to the Trustee Council by November 22, 1993. All other projects and the final project report will be submitted by April 15, 1994. The Trustee Council will decide which, if any, of the restoration projects identified will be funded.

Project Description

D. Location

Meetings are being held in all communities in PWS as well as Seward and Anchorage. Additionally, meetings with individuals or groups are being held at locations most convenient to them, although typically in Anchorage.

E. Technical Support

Technical support will be needed to input information from the recreation questionnaire and analyze questionnaire data. Support will also be needed from ADNR to develop working maps of PWS for use in meetings and for identifying and mapping potential project locations.

F. Contracts

Contractual funds will be utilized to provide charter aircraft for travel to villages in PWS, meeting space for the November 5-6 workshop, and a meeting facilitator for the workshop.

SCHEDULES

A schedule of the major milestones for project tasks, public meetings, analysis, and final reports is presented in Table 1. Initial data collection began in April, 1993. Meetings with the different interest groups began in June and will continue through September. A list of all the projects proposed by the public will be prepared for the November 5-6, 1993 workshop. Complete project descriptions for the best projects identified will be submitted by November 22, 1993 for the FY 94 Work Plan. All other projects identified will be listed and prioritized as a part of the project report, April 15, 1993. Investigations of possible special designations began in April, 1993. A preliminary draft of possible designations will be available the end of September, 1993. Complete findings and recommendations will be a part of the project report. Development of goals and objectives began in June, 1993. A statement of injury to the recreation resource and services will be completed in September, 1993. A summary of the findings related to injury and any restoration goals and objectives will be included in the project report.

Table 1. Project Schedule.

Date	Accomplishment
	Complete major collection of data, projects submitted, surveys, etc. Additional data collection will be ongoing as necessary.
June 1, 1993	Begin meetings with interest groups. Continues through October, 1993 Currently scheduled:
	July 21-23 Valdez
	July 27 Chenega Bay
	August 6 Seward
	August 10-11 Cordova
	August 16 Whittier
	August 24 Tatitlek
September 1, 1993	Draft statement of injury to recreation resources and services of PWS
September 30, 1993	Final statement of injury to recreation resources and services of PWS
October 1, 1993	Draft management goal and objectives for PWS
October 30, 1993	Preliminary list of project proposals and descriptions
November 5-6, 1993	Workshop with all interested groups
November 22, 1993	Project descriptions for FY94 Work Plan
February 15, 1994	Draft Project Report for peer review
April 15, 1994	Final Project Report

EXISTING AGENCY PROGRAM

The State of Alaska Department of Natural Resources has been funded for \$4.75M of the criminal settlement to conduct recreation restoration activities within the spill area. The Chugach National Forest manages PWS with primary management emphasis on recreation and fish and wildlife management. Cabin maintenance, trail maintenance and development, mooring buoy placement, and cultural resources site identification, protection, and interpretation are some of the major tasks performed each year. The Forest Service has allocated approximately \$90K per year for the management of recreation within PWS. Additional funds have been allocated for fish and wildlife management. The Chugach

Project Description

onal Forest Land Management Plan is scheduled for revision starting in 1995, and goals and objectives identified by this project may be adopted in the revised plan.

ENVIRONMENTAL COMPLIANCE/PERMIT/COORDINATION STATUS

This project is categorically exempt from formal documentation in an environmental assessment or environmental impact statement under Forest Service regulations [FSH 1909.15 31.1a(3)]. No other permits are required. No ground disturbing activity is proposed. This project is for recreation implementation development. Projects developed from this project may require environmental analysis or permits before implementation.

PERFORMANCE MONITORING

Products provided from this project include:

1. Complete project descriptions for the most desirable recreation restoration projects for the FY 1994 Work Plan, based on the public involvement process being used.
2. A final project report describing the process followed, contacts made, workshop results, possible special designations, and a listing of all projects identified by priority.
3. A statement of injury to the recreation resource and services of PWS.

FY94 BUDGET (\$K)

	USFS	ADNR	TOTAL
Personnel	27.4	32.5	59.9
Travel	1.9	1.9	3.8
Contractual	6.8	7.0	13.8
Commodities	3.5	0.2	3.7
Equipment	0.0	0.0	0.0
Capital Outlay	<u>0.0</u>	<u>0.0</u>	<u>0.0</u>
Sub-total	39.6	41.6	81.2
General Administration	<u>4.6</u>	<u>5.4</u>	<u>10.0</u>
Project Total	44.2	47.0	91.2
NEPA Compliance	0.0		

EXXON VALDEZ TRUSTEE COUNCIL
FY 94 DETAILED PROJECT DESCRIPTION

A. COVER PAGE

Project title: Harbor Seal and Sea Otter Co-op Subsistence Harvest Assistance

Project ID number: 94244

Project type: Subsistence

Name of project leader(s): Rita A. Miraglia

Lead agency: Alaska Department of Fish and Game

Cooperating agencies: None

Cost of project/FY 94: \$54,500

Cost of project/FY 95: \$54,500

Cost of Project/FY 96 and beyond: \$54,500

Project Start-up/Completion Dates: 4/1/94 through 9/30/94

Geographic area of project: Prince William Sound and the Lower Kenai Peninsula

Name of project leader: Rita A. Miraglia

Name of lead agency project manager: Dr. Joseph R. Sullivan

B. INTRODUCTION

The populations of harbor seals and sea otters in Prince William Sound and adjacent waters were injured as a result of the Exxon Valdez oil spill. The U. S. Fish and Wildlife Service estimates that between 3,500 and 5,500 sea otters were killed by oil in the first months after the spill, and sea otters were still being injured by oil in the environment three years later. The case for a population level oil spill injury to harbor seals is less clear. Harbor seal populations throughout the Gulf of Alaska are known to have been in decline before the oil spill. This decline has continued, but it is difficult to determine how much, if any of it is due to the effects of oil in the environment. However, it is known that harbor seals were exposed to Exxon Valdez crude oil in Prince William Sound. They suffered some direct mortality although the number of harbor seals killed is unknown. Harbor seals also suffered sub-lethal effects, including corneal damage, nerve damage, and brain lesions.

Many subsistence hunters within the spill area, concerned about the decline they have observed in the numbers of harbor seals and sea otters, have voluntarily reduced their take of these species in an effort to help their recovery. However, at present, there is no mechanism in place to evaluate the effectiveness of these efforts.

Some data are available on marine mammal harvests in the spill area. The Division of Subsistence, Alaska Department of Fish and Game has collected information on the numbers of harbor seals and sea otters harvested by subsistence users living in several communities in the spill region for both pre-and post-spill years. In 1993, the Division of Subsistence, in cooperation with the National Marine Fisheries Service and Ruralcap, also undertook a project to collect more detailed information on the timing and composition of subsistence harvests of harbor seals and sea lions (but not sea otters), including figures for those animals struck and lost. The U.S. Fish and Wildlife Service runs a sea otter tagging program, which gathers information on sea otter harvests, including the location where animals are taken.

There is also some information available on harbor seal and sea otter populations in the region. The Division of Wildlife Conservation, Alaska Department of Fish and Game, working with the National Marine Fisheries Service has conducted a count of harbor seals in both the oiled and unoled areas of Prince William Sound, along with other research aimed at assessing the health of the harbor seals (restoration project number 93046). The U.S. Fish and Wildlife Service has continued to monitor the recovery of sea otters in oiled areas, by determining their abundance, distribution and mortality (restoration project number 93043).

We propose: 1) to compile all of the available information; 2) gather additional data as needed; 3) analyze and interpret the data, in cooperation with the appropriate agencies and native groups; 4) provide an interpretation of the data regarding

harbor seal and sea otter population dynamics, trends, and harvests, and their relationships, to subsistence hunters; and 5) if shown to be appropriate by a review of the data, produce a set of recommendations to subsistence users to guide those who want to voluntarily change their harvesting practices to help these two species recover.

This project constitutes a step towards involving subsistence hunters in the resource management process, and may lead to an on-going exchange of information and consensus building with regard to the management of harbor seals.

C. PROJECT DESCRIPTION

1. Resources and/or Associated Services:

The goal of the project is to assess the available data about the relationship between population dynamics and trends and subsistence harvests of harbor seals and sea otters, and to work cooperatively with subsistence hunters to assess the relationship between these factors and the recovery of these species.

2. Relation to Other Damage Assessment/Restoration Work:

This project will incorporate information on the numbers, distribution and degree of recovery of the populations of harbor seals and sea otters from restoration projects 94064 (Harbor Seal Habitat Use and Monitoring) and 94246 (Sea Otter Recovery Monitoring Project). It will also provide information on seal and sea otter harvest in Prince William Sound and the Lower Kenai Peninsula to help the marine mammal researchers to evaluate the impacts, if any, of the harvest on the recovery of those populations.

3. Objectives:

An interpretation of the available information about the relationship between harbor seal and sea otter population dynamics and trends and subsistence harvests to provide to subsistence hunters of these species by September 1994. If a review of the data indicates changes in harvest practices would aid in the recovery of these species, a set of recommendations will be produced to guide those subsistence users who want to voluntarily change their harvesting practices to help these two species recover.

4. Methods:

The project will involve compiling information from a variety of sources. Sea otter tagging data collected by the U.S. Fish and Wildlife Service will be used to estimate both the number of sea otters taken, and the locations in which they

were hunted. The Division of Subsistence is already administering a survey in the oil spill impacted communities to gather information on the numbers of harbor seals harvested, including a breakdown by life stage and sex of the animal, and an estimate of the number struck and lost. A section would need to be added to the questionnaire on location of harvest for this species. This would also increase the length of time it takes to conduct each interview. Information collected by the Alaska Department of Fish and Game, the U.S. Fish and Wildlife Service, and the National Marine Fisheries Service on the biology and population characteristics of harbor seals and sea otters in the oil spill impact area will also be used. The Division would need to contract with an organization such as the Alaska Sea Otter Commission or Ruralcap to assist in the interpretation of the biological and population data, and the potential effects of the harvest on the health of the populations, as such interpretation is outside of our expertise.

Following the compilation and analysis of the data, an ad hoc committee would be convened to evaluate the accumulated information, provide an interpretation of the data to subsistence users, and if appropriate make recommendations regarding changes in harvest practices. Such a committee would be composed of representatives of appropriate agencies, including the Alaska Department of Fish and Game, the US Fish and Wildlife Service, and the National Oceanic and Atmospheric Administration, and native organizations, including the Alaska Sea Otter Commission, Ruralcap, the Chugach Regional Resources Commission, and the village and traditional councils of the area. The interpretations as well as any recommendations of the ad hoc group would be presented to subsistence users both in an informational newsletter, and in community meetings. Any changes to the harvest would be voluntary, as the ad hoc group would have no authority to compel any changes.

5. Location:

The study area will include Prince William Sound and the lower Kenai Peninsula.

6. Technical Support:

The project will not require any technical support as defined in the instructions for this document.

7. Contracts:

The Division of Subsistence will need to contract out for someone to assist in the analysis and interpretation of the biological and population data. This kind of work is outside the expertise of the staff of the Division of Subsistence, and the Division of Wildlife Conservation will not have any personnel available for this task. We would prefer to contract with one or more native organization(s), such as Ruralcap or the Alaska Sea Otter Commission for this service, because their ties to the native communities would be of benefit to the project.

D. SCHEDULES

MAY 1994:

Finish collection of harvest information for 1993
Collect and compile traditional knowledge from hunters regarding harbor seal and sea otter populations
Identify and evaluate alternative data sources such as information from fur buyers

JUNE 1994

Meeting between marine mammal biologists and staff of the Division of Subsistence

JULY 1994:

Finish compilation of biological and population data

AUGUST 1994:

Convene ad hoc group to evaluate data

SEPTEMBER 1994:

Provide data interpretations and any recommendations to subsistence harvesters
Produce informational newsletter
Hold community meetings

A second year of harvest monitoring and evaluation of new biological and population data will be needed to complete the project.

E. EXISTING AGENCY PROGRAM

This project will take advantage of several existing programs, both within the Alaska Department of Fish and Game, and other agencies. These other programs are described above.

F. ENVIRONMENTAL COMPLIANCE/PERMIT/COORDINATION STATUS

Since there is no field work component to this project, it is categorically excluded under NEPA guidelines.

G. PERFORMANCE MONITORING

A database will be compiled for this project. The ad hoc group will develop a set of data interpretations and, if appropriate, recommendations for subsistence users of the two species, and an informational newsletter will be produced. A report will be completed at the end of each project year.

H. COORDINATION OF INTEGRATED RESEARCH EFFORT

This project will incorporate information on the numbers, distribution and degree of recovery of the populations of harbor seals and sea otters from restoration projects 94064 (Harbor Seal Habitat Use and Monitoring) and 94246 (Sea Otter Recovery Monitoring Project). It will also provide information on seal and sea otter harvest in Prince William Sound and the Lower Kenai Peninsula to help the marine mammal researchers and subsistence users to cooperatively discuss possible relationships between subsistence harvests and the recovery of these populations.

I. PUBLIC PROCESS

This project will incorporate public input from community meetings. Representatives of the communities of Prince William Sound and the Lower Kenai Peninsula will be included as members of the ad hoc body evaluating the data. Any findings of the ad hoc body will be disseminated to the communities by means of an informational newsletter as well as in community meetings.

J. PERSONNEL QUALIFICATIONS

James Fall: Dr. Fall is the regional program manager for the Division of Subsistence, Alaska Department of Fish and Game, for southcentral and southwest Alaska. He has held this position since 1981. Since 1989, he has supervised the division's oil spill response and research program. Also, he has served as the department's representative on the Oil Spill Health Task Force. Dr. Fall has written several articles and reports on the effects of the *Exxon Valdez* oil spill on subsistence activities and harvests, based upon division research.

Rita Miraglia: Ms Miraglia has served as the oil spill coordinator for the Division of Subsistence since 1990. As such, she has organized and participated in the subsistence resource collection and testing programs of 1990 and 1991. She has also been the lead communicator of study findings to communities through organizing community meetings and writing newsletters. She has also assisted the Oil Spill Health Task Force's activities.

Craig Mishler: Dr. Mishler has been a Subsistence Resource Specialist with the Division of Subsistence since 1989, with primary responsibility for Kodiak Island. He is project leader for the division's seal and sea lion harvest monitoring program.

Jody Seitz: Ms Seitz has worked as a Subsistence Resource Specialist with the Division of Subsistence since 1989, with responsibility for Prince William Sound communities since 1991. She has participated extensively in Division of Subsistence research projects in these communities, including the collection of information about subsistence harvests after the oil spill.

K. BUDGET

EXXON VALDEZ TRUSTEE COUNCIL
1994 Federal Fiscal Year Project Budget
October 1, 1993 - September 30, 1994

Project Description: Seal & Otter Co-op Subsistence Harvest Assistance - The purpose of this project is to gather information on harbor seal health, biology and demographics, as well as figures on subsistence harvests of these two species in Prince William Sound and the lower Kenai Peninsula; an ad hoc body of representatives of agencies and native organizations will be assembled to evaluate this information and make recommendations for changes in harvesting, if warranted, to help these species recover in the wake of EVOS.

Budget Category:	1993 Project No. Authorized FFY 93	'93 Report/ '94 Interim* FFY 94	Remaining Cost** FFY 94	Total FFY 94	FFY 95	Comment
Personnel	\$0.0	\$0.0	\$32.0	\$32.0	\$32.0	
Travel	\$0.0	\$0.0	\$5.0	\$5.0	\$5.0	
Contractual	\$0.0	\$0.0	\$10.0	\$10.0	\$10.0	
Commodities	\$0.0	\$0.0	\$2.0	\$2.0	\$0.8	
Equipment	\$0.0	\$0.0	\$0.0	\$0.0	\$1.2	
Capital Outlay	\$0.0	\$0.0	\$0.0	\$0.0	\$0.0	
Subtotal	\$0.0	\$0.0	\$49.0	\$49.0	\$49.0	
General Administration	\$0.0	\$0.0	\$5.5	\$5.5	\$5.5	
Project Total	\$0.0	\$0.0	\$54.5	\$54.5	\$54.5	
Full-time Equivalents (FTE)	0.0	0.0	0.5	0.5	0.5	
Dollar amounts are shown in thousands of dollars.						
Budget Year Proposed Personnel:		Reprt/Intrm Months	Reprt/Intrm Cost	Remaining Months	Remaining Cost	
Position Description						
Subsistence Resource Specialist II		0.0	\$0.0	2.0	\$9.4	
Subsistence Resource Specialist II		0.0	\$0.0	3.0	\$13.7	
Research Analyst		0.0	\$0.0	1.0	\$5.5	
Program Manager		0.0	\$0.0	0.5	\$3.4	
Personnel Total		0.0	\$0.0	6.5	\$32.0	
NEPA Cost:						\$0.0
*Oct 1, 1993 - Jan 31, 1994						
**Feb 1, 1994 - Sep 30, 1994						

07/14/93

1994

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Project Number: 94244
Project Title: Seal & Otter Co-op Subsistence Harvest Assistance
Agency: AK Dept. of Fish & Game

FORM 2A
PROJECT
DETAIL

EXXON VALDEZ TRUSTEE COUNCIL
 1994 Federal Fiscal Year Project Budget
 October 1, 1993 - September 30, 1994

Travel:	Reprt/Intrm	Remaining
Visits to the various native communities to collect information and report recommendations		
Anchorage to Chenega (2 trips - air fare \$900/trip)	\$0.0	\$1.8
Anchorage to Tatitlek (2 trips - air fare \$550/trip)	\$0.0	\$1.1
Anchorage to Valdez (2 trips - air fare \$160/trip)	\$0.0	\$0.3
Anchorage to Cordova (2 trips - air fare \$205/trip)	\$0.0	\$0.4
Per diem (13 days @ \$105/day)	\$0.0	\$1.4
Travel Total	\$0.0	\$5.0
Contractual:		
Contract with wildlife biologist to analyze the data and submit a final report (This contract will be awarded through the competitive bid process for the compilation and analysis of the available seal and sea otter harvest and population data.)	\$0.0	\$10.0
Contractual Total	\$0.0	\$10.0

07/14/93

1994

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Project Number: 94244
 Project Title: Seal & Otter Co-op Subsistence Harvest Assistance
 Agency: AK Dept. of Fish & Game

FORM 2B
 PROJECT
 DETAIL

Commodities:

Reprt/Intrm	Remaining
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\$0.0	\$0.8
\$0.0	\$1.2

Commodities Total	\$0.0	\$2.0
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Equipment:

Equipment Total	\$0.0	\$0.0
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**NO DETAILED PROJECT DESCRIPTION REQUIRED --
PROJECT IS A CONTINUATION OF
PREVIOUSLY APPROVED WORK**

BRIEF PROJECT DESCRIPTION FOLLOWS

EXXON VALDEZ OIL SPILL PROJECT DESCRIPTION

Title: Sea Otter Recovery Monitoring

Project Number: 94246

Lead Agency: DOI-FWS

Cooperating Agency: None

Cost of Project, FY94: \$418.7K

Cost of Project, FY95: \$70.6K

Project Startup Date: April 1994

Duration: 1 year

Geographic Area: Prince William Sound

INTRODUCTION

Sea otters (*Enhydra lutris*) are a well-known marine mammal species in Alaska. They historically occurred throughout coastal waters of the Pacific, but as a result of fur harvests in the 18th and 19th centuries, they came close to extinction. They have since increased in abundance and distribution, and presently are found in most coastal areas of southern Alaska. Sea otters prey on a variety of invertebrate species, including mussels, clams, crabs, and sea urchins.

Acute losses of sea otters associated with the Exxon Valdez oil spill probably ranged from 3,500 to 5,000 animals. Natural Resource Damage Assessment studies on (1) sea otter abundance in Prince William Sound (PWS), (2) age distribution of sea otter carcasses recovered in oiled areas, and (3) first-year survival of juvenile sea otters in PWS suggested that oil-related injury to the population persisted through 1991.

Data collection in 1992 was limited; however, age distributions of carcasses recovered in 1992 suggested mortality was returning to a pre-spill pattern. In addition, a juvenile survival study conducted by USFWS in 1992-93 indicates improved rates of first year survival compared to 1990-91. Analyses of blood of sea otters caught in oiled areas in 1992, however, showed that approximately 25% of the adult otters have significant elevations in serum enzymes associated with liver and kidney damage. Restoration studies underway in 1993-94 will supply additional information on population size and distribution. The restoration project proposed herein for 1994-1995 will monitor the population to assess whether or not there is evidence of recovery and obtain data to evaluate condition of sea otters and possible pathologies that may be contributing to continued injury in the population.

PROJECT DESCRIPTION

A. Resources and/or Associated Services

This project will monitor sea otters.

B. Objectives

1. Monitor Spring/Summer 1994 abundance and distribution of sea otters in PWS for comparison with previously collected data.
2. Monitor 1994 mortality trends in sea otters such as age class, gender, and location.
3. Investigate pathological conditions of sea otters in PWS. Determine if the pathological conditions observed in 1993 still exist.

C. Methods

1. Abundance and distribution of sea otters in PWS will be monitored through application of aerial surveys developed by USFWS (with partial support of restoration funds) in 1991-1993.
2. Mortality of sea otters will be evaluated by examining the age distribution of beach-cast carcasses retrieved on annual beach walks. Sea otter age will be determined, as in the past, by tooth cementum analysis.
3. Pathological conditions of sea otters will be examined by (1) analysis of hematology and serum chemistry in blood samples collected from live sea otters and (2) gross examination of carcasses and histologic examination of tissue samples collected from carcasses of sea otters taken by subsistence hunters. No alternative methods exist for collection of data.

D. Location

Field work will take place in PWS. Data entry, analysis, and report preparation will take place in Anchorage.

E. Technical Support

None needed.

Contracts

Contracts of \$0.4 to \$0.8K each will be required for determination of age from teeth, for blood analysis, and for necropsies of any intact carcasses that may be recovered.

SCHEDULES

1. Field work - April 15, 1994 to September 15, 1994:

April/May - Beach walks for carcass retrieval.

July - Blood collection from live sea otters.

August - Aerial surveys for abundance and distribution monitoring.

by September - Collection of carcasses from subsistence takes will depend on hunters but we anticipate obtaining samples by September.
2. Analysis of biological samples (blood, teeth, histopathology) - July to October, 1994.
3. Data entry and analysis - August to November, 1994.

Draft reports will be completed by January 16, 1995.
5. Final reports will be completed by March 31, 1995.

EXISTING AGENCY PROGRAM

The USFWS covers salary for two investigators during their contributions to this project (primarily the aerial surveys) - total estimated agency contribution is \$14.9K.

The USFWS will be radiotracking juvenile sea otters in PWS on a bi-weekly basis until June 1994, to measure second year survival rates and movements.

ENVIRONMENTAL COMPLIANCE/PERMIT/COORDINATION STATUS

A Marine Mammal Permit is required for handling sea otters and collection of blood; a permit which covers this work has already been issued. Other components of the study involve collection of samples from sea otter carcasses and do not require an additional permit. The aerial survey relies on observations from a small airplane and is non-intrusive.

Based on a review of CEQ regulation 40 CFR 1500-1508, this study has been determined to be categorically exempt from the requirements of NEPA, in accordance with 40 CFR 1508.4.

PERFORMANCE MONITORING

Final reports provided by March 31, 1995, will demonstrate that project objectives have been met.

FY94 BUDGET (\$K)

USFWS

Personnel	255.0
Travel	11.4
Contractual	88.4
Commodities	19.5
Equipment	0.0
Capital Outlay	<u>0.0</u>

Subtotal	374.3
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General Administration	<u>44.4</u>
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Project Total	418.7
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NEPA Compliance	0.0
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EXXON VALDEZ TRUSTEE COUNCIL
FY 94 DETAILED PROJECT DESCRIPTION

Project title: Kenai River Sockeye Salmon Restoration

Project ID number: 94255

Project type: General Restoration

Name of project leaders: Ken Tarbox, Lisa W. Seeb, James E. Seeb

Lead agency: Alaska Department of Fish and Game
(ADF&G)

Cooperating agencies: None

Cost of project/FY 94: \$406.1K

Cost of project/FY 95: \$350.0K

Cost of Project/FY 96 and beyond: \$300.0K

Project Start-up/Completion Dates: January 1992 and to continue five years

Geographic area of project: Upper Cook Inlet

Project leaders:

Linda Brannigan for Ken Tarbox 3/14/94
Ken Tarbox Date

Lisa W. Seeb 3/14/94
Lisa W. Seeb Date

Lisa Seeb for James E. Seeb 3/14/94
James E. Seeb Date

Agency project manager:

Joseph R. Sullivan 3/14/94
Joe Sullivan Date

B. INTRODUCTION

Sockeye salmon (*Oncorhynchus nerka*) which spawn in the Kenai River system were injured by the Exxon Valdez oil spill (EVOS). Greatly reduced fishing time in Upper Cook Inlet (UCI) due to EVOS caused sockeye salmon spawning escapement levels in the Kenai River to exceed that desired by three times. The biological impact of EVOS on Kenai River sockeye salmon stocks may be one of the most serious documented. Data collected by NRDA Fish/Shellfish Study 27, *Sockeye Salmon Overescapement*, indicated greatly reduced survival of juvenile sockeye salmon during the winter-spring rearing period beginning with the 1989 parent year. The extremely high escapement may have initially produced more rearing juvenile sockeye salmon than could be supported by nursery lake productivity.

In general, when rearing salmon abundance greatly exceeds lake carrying capacity, the species and size composition of prey resources are altered which affects all trophic levels. Because of such changes, juvenile sockeye growth is reduced, freshwater mortality is increased, greater proportions of fry remain in the lake for another year of rearing, and smolt condition is reduced and marine mortality is increased.

Limiting sockeye salmon fry production by closely regulating the number of spawning adults may be the only way to restore the productivity of these rearing areas. Sockeye smolt outmigrations in the Kenai River were severely reduced in 1991 (1989 parent year) and have continued to decline through 1993. The number of adult sockeye salmon returning from the 1989 overescapement in the Kenai River is expected to be low. Starting in 1994, a dramatic reduction or complete elimination of Kenai River sockeye salmon harvests may be necessary to ensure even minimally adequate escapements.

Sockeye salmon harvested from the mixed-stock fishery of Cook Inlet include fish from the Kenai, Kasilof, and Susitna Rivers. In order to effectively manage the harvest of EVOS-damaged stocks, Restoration Science Studies R53/R59/93012/93015 were implemented in 1992 and 1993. These studies use Genetic Stock Identification (GSI) techniques to identify Kenai River stocks in mixed stock Cook Inlet fisheries. Area managers will use this information to modify fishing areas and openings in order to facilitate the harvest of surplus Kasilof and Susitna River stocks while protecting the EVOS-damaged Kenai River stocks. Project 94255 is the continuation of these projects through fiscal 1994.

Attempts to use stock identification to manage harvest of Cook Inlet sockeye salmon in the past have relied on scale growth patterns. However, the accuracy of the scale technique alone has not been reliable, and it is insufficient to permit the in-season protection of the EVOS-damaged Kenai River stocks. GSI techniques rely on genetic variation to discriminate between populations of organisms. This method has recently been applied in other areas as an in-season fisheries management tool, and it has proven to be extremely effective for allocating and adjusting the harvest of fish stocks intercepted in mixed-stock fisheries such as those that occur in Cook Inlet. Once a data base has been established, GSI techniques should provide a mechanism for in-season management on a stock-specific basis. This will allow managers to control the harvest of

Kenai River sockeye salmon and facilitate their recovery.

A pilot study was conducted during 1993, prior to the return of the first EVOS-impacted stocks anticipated in 1994 (age-5 sockeye salmon from the 1989 parent year). Two fishery samplings were completed using the genetic baseline collected during the 1992 field season. In both cases the laboratory and statistical analysis were completed within 48 hours. Preliminary estimates indicate that this type of information can be sufficiently precise to inform fishery managers on current stock composition in Cook Inlet.

C. PROJECT DESCRIPTION

1. Resources and/or Associate Services:

Restoration of Kenai River sockeye stocks will benefit subsistence, sport, and commercial fishermen in coastal communities throughout Cook Inlet, from Homer north through Anchorage to Tyonek. In 1992 nearly 10,000 families obtained subsistence permits to harvest salmon in UCI, most targeting Kenai River sockeye salmon. The most recent statistics indicate that nearly 100,000 sport anglers fished the Kenai River for salmon in 1990, spending \$38 million in 1986 dollars. Forty percent of those anglers were from out of state. Of the 1,323 permits licensed to commercial fish in UCI, 80% are fished by state residents with the remaining predominately from Pacific Coast states. Average ex-vessel value (1987-1991) of the UCI commercial salmon harvest was \$67.8 million.

2. Relation to Other Damage Assessment/Restoration Work:

Restoration project 94258 (Sockeye Overescapement) will be collecting samples of juvenile salmon (fry and smolt) for genetic analysis. In addition, results from project 94258 will be incorporated with results from this investigation to formulate an over-all restoration plan for Kenai River sockeye salmon.

3. Objectives:

The goal of this project is to restore Kenai River sockeye salmon injured by the oil spill. This will be accomplished through improved stock assessment capabilities, more accurate regulation of spawning levels, and modification to human use. The specific objectives are to:

1. Obtain baseline allozyme genetic data (during 1992-1995) from all significant spawning stocks contributing to mixed-stock harvests of sockeye salmon in Cook Inlet.
2. Use Genetic Stock Identification (GSI) algorithms to estimate the proportion of Kenai River stocks in mixed stock fisheries so that managers may modify area and time of harvest in order to protect these damaged stocks while targeting surplus Kasilof River and Susitna River stocks. Genetic data will be obtained from samplings of the various mixed-stock fisheries occurring in 1994-1996. Stocks composition estimates will be provided within 48 hours

post-fishery. In addition, GSI data may be used to identify the contributions of Kenai River sub-stocks in samples collected during Restoration Project 94258 (Kenai River Sockeye Overescapement).

3. Investigate the added utility of DNA-level markers to discriminate among Cook Inlet populations.
4. Provide more accurate estimates of abundance of Kenai River sockeye salmon within Cook Inlet through hydroacoustic assessment techniques.

4. Methods:

Stock Identification:

We will continue to develop a comprehensive genetic database of sockeye salmon stocks in Cook Inlet. In 1992 we collected baseline genetic data using allozyme analyses from 28 subpopulations from Cook Inlet including the Kenai, Kasilof, and Susitna Rivers. Additional sockeye salmon were collected from approximately 34 baseline subpopulations in 1993. An additional ten sites will be chosen each year (1994-1995) to supplement or confirm those previously sampled. Final selection of sample sites will depend on GSI results from the previous year. Target sample sizes for allozyme baseline collections will be 100 individuals to adequately characterize spawning populations (Allendorf and Phelps 1981, Waples 1990).

Mixed stock fishery samples will be collected from selected drift fisheries openings occurring during the July fisheries (maximum of two; 1994-1996). In addition, mixture samples of Kenai River smolt and fry will be collected during Restoration Project 94258. Mixed stock sample sizes will be set at 400 to minimize the confidence intervals surrounding the estimates (Pella and Milner 1987).

Muscle, liver, eye, and heart will be dissected from freshly killed individuals. Tissues will be placed in labeled cryovials and transferred into liquid nitrogen (Appendix A, B). Tissues from baseline collections will be stored on liquid nitrogen until transferred to -80°C storage in Soldotna or Anchorage. Soldotna samples will be transferred to the Anchorage laboratory on dry ice or liquid nitrogen and again placed in -80°C storage where they will remain until laboratory analysis.

Allozyme electrophoretic data (Utter et al. 1987, Seeb et al. 1987) will be collected for the loci identified in during year one and two of the study (Table 1). An extensive allozyme screening was undertaken to maximize the potential number of available gene markers. A total of 84 allozyme loci were resolved (Table 1). Of the resolvable loci, 42 were polymorphic in at least one individual (*sAAT-1,2*; *mAAT-1*; *mAAT-2*; *MAH-1,2*; *MAH-4*; *SAH*; *ALAT*; *CK-A2*; *CK-C1*; *CK-C2*; *CK-B*; *FDH*; *GAPDH-2*; *GDA-1*; *G3PDH-1,2*; *GPI-A*; *GPI-B1,2*; *mIDHP-1*; *sIDHP-1*; *LDH-A2*; *LDH-B1*; *LDH-B2*; *sMDH-A1,2*; *sMDH-B1,2*; *mMEP*; *MPI*; *PEPA*; *PEPB-1*; *PEPD-1*; *PEPLT*; *PGDH*; *PGM-1*; *PGM-2*; *TPI-1,2*; *TPI-4*)

All gel scoring will be conducted on-line to ensure rapid turnaround, complete

documentation, and immediate availability of summary statistics. Allozyme techniques will follow those of Harris and Hopkinson (1976), May et al. (1979), and Aebersold et al. (1987); nomenclature rules will follow the American Fisheries Society standard (Shaklee et al. 1990). A photographic record of each gel will be made.

Genotypic and allelic frequency estimates will be calculated for each baseline and mixed-stock sample at every locus. Genetic distance measures (Nei 1978), which summarize multi-locus data into a single number, will also be calculated between all pairs of spawning locations. These values will be used to construct branching diagrams using numerical taxonomic techniques (UPGMA) which provide a representation of overall phenetic similarity. A neighbor-joining tree (Saitou and Nei 1987) will be constructed to provide a phylogenetic tree relating the populations. Chi-square goodness-of-fit to Hardy-Weinberg equilibrium will be performed to test for random mating within each population.

Homogeneity of allelic frequencies among the various collections will be tested using a log-likelihood ratio analysis (G-statistic; Smouse and Ward 1978; $\alpha = 0.01$; Cooper 1968). Rejection of the null hypothesis of homogeneity is indicative of discrete spawning populations. The total gene frequency dispersion at each locus will be subdivided into within-and among-river system components in a hierarchical fashion. Hierarchical levels will be organized to test for homogeneity of (1) within drainages of the systems; (2) among drainages within river systems; and (3) among river systems. The likelihood analysis will use the computational formula of Sokal and Rohlf (1981). This statistic is distributed approximately as the chi-square statistic with $(\text{no. of alleles} - 1) \times (\text{no. of region} - 1) = (\text{degrees of freedom})$. The likelihood values (G) can be summed over all loci to obtain a total value at each level of analysis.

Stock contribution to mixed fishery samples will be estimated using a conditional maximum likelihood program (GIRLSEM) developed by National Marine Fisheries Service (NMFS; Pella and Milner 1987, Masuda et al. 1991; see Appendix C). The precision of the stock composition estimates will be determined by bootstrap resampling (Efron and Tibshirani 1986). In bootstrapping, individuals of the stock and mixture samples are randomly resampled with replacement to obtain new samples equal in size to the original samples. Standard errors of stock composition estimates due to sampling errors in the stock and mixture samples can be estimated from the standard errors of composition estimates over resamplings of the bootstrap. Approximately 100 bootstrap resamplings should provide sufficiently accurate estimates of standard error (Masuda et al. 1991). Accuracy graphs will be obtained by constructing simulated samples of mixtures with specific stock proportions and then by bootstrap resampling the baseline to obtain estimates of stock proportions. This same type of simulation will be used to evaluate the effect of mixture sample size on the accuracy and precision of the stock composition estimates and will be used to adjust mixture sample size in succeeding years.

Considerable progress has been made in developing the analytical and computational techniques to rapidly provide fishery estimates for in-season management. Development of a comprehensive package of genetic analysis programs Windows applications (Microsoft 1991) has begun and includes the following components: (1) an on-line gel scoring program providing extensive documentation of results and error checking

capability; (2) a set of genetic analysis programs providing allele frequency estimates, heterogeneity analyses, and fit to expected genetic models; and (3) revised input into the GIRLS algorithm (Masuda et al. 1991) to allow rapid fishery estimates and a flexible method to conduct multiple simulation studies. The object oriented genetics applications will work synchronously within the Windows environment to provide a user friendly interface for data input and complicated analyses so that the geneticists can make a fast turn-around from field samples to fishery estimates. Functional versions (beta) of these programs are now in use. The allozyme data will be analyzed using the genetic analysis program, BIOSYS-1 (Swofford and Selander 1981) and NTSYS-pc (Rohlf 1993). Fishery composition estimates will be available within 48 hours following the fishery so that management decisions can be based on the actual composition of the fisheries.

A pilot study using DNA techniques will be conducted on a subset of the collected allozyme baseline samples. DNA will be extracted from liver and heart tissue (Chapman and Brown 1990; Bermingham et al. 1991) using standard protocols of Proteinase-K and RNase-A digestion, phenol/chloroform extraction and ethanol precipitation (Sambrook et al. 1989). After extraction, the purified 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 selections for PCR will include the universal primers for the mitochondrial DNA (mtDNA) cytochrome-b gene (Kocher et al. 1989) and primers for the mtDNA ND5\6 and ND3\4L\4 regions (Cronin et al. 1993). These regions have proven useful in other stock identification studies within the genus *Oncorhynchus* (Cronin et al. 1993). The amplified mtDNA regions will then be digested with up to 20 restriction enzymes and the resulting fragments will be separated on agarose gels. The restriction fragment length polymorphisms (RFLPs) produced will then be visualized under ultraviolet light, and a photographic record will be made of each gel. Since genes which are encoded by the mitochondrial genome are inherited as a single unit (i.e., analogous to linked loci), the restriction sites detected for each enzyme, for all regions examined, will be pooled together as composite haplotypes. The frequencies and distributions of these composite haplotypes will then be used to examine the structure of salmon populations.

Nucleotide (d) and haplotype (h) diversity measures (Nei 1987) will also be calculated for all populations using the restriction enzyme analysis package (REAP) of McElroy et al. (1992). These measures estimate the number of nucleotide substitutions per site between DNA sequences (i.e., sequence divergence) and the amount of DNA polymorphism within populations, respectively. These values will then be used to calculate an overall genetic distance (Nei 1978) between populations, which in turn, will be used to generate a branching diagram using the Fitch and Margoliash (1967) least-squares algorithm in the *PHYLIP* (Felsenstein 1993) package. This dendrogram will depict relationships among the populations. Stability of this dendrogram will be tested using the bootstrap method of Felsenstein (1985).

Offshore Test Fish Program:

The sockeye salmon total run to UCI has been estimated early during the season by test fishing between Anchor River and Red River delta. Northward migrating sockeye salmon are captured with a drift gill net at a series of stations. Salmon are identified to species

and sex, and length measurements are recorded. Estimates of total sockeye salmon return are made several times during the season by estimating expected total test fishery catch per unit of effort for the season and catchability of sockeye salmon in the test fishery. Analysis of historical data has indicated that existing sampling effort and catch has not been proportional to abundance. To assess run size more accurately, additional sampling effort will be added to the existing program.

In 1992 and 1993 hydroacoustic equipment and techniques were tested in UCI offshore waters (Thorne and Salomone 1993; Thorne 1994). Results of this work indicated that hydroacoustic techniques could detect salmon and provide a population estimate for "in season" management use. However, the primary limitation identified in the study was limitations (signal/noise ratio) of the hydroacoustic gear due to rough sea conditions or shallow water in the northern portion of UCI.

In 1994 two abundance estimates will be made using the techniques developed in 1992 and 1993. Examination of the data set indicated that a minimum of 12 orthogonal transects sampled over 48 hours within Cook Inlet would be needed to provide a useable estimate of adult salmon abundance. Therefore, a minimum of 12 transects will be completed during each survey. Exact timing of the survey will be determined during the commercial fishing season to meet commercial fishery management objectives.

5. Location:

Upper Cook Inlet, north of a line from Anchor Point to the Red River Delta - Field work will be inlet wide and based out of Soldotna; lab and a portion of the data analyses will be conducted in Anchorage.

6. Technical Support:

Administrative support is provided by the Administrative, Habitat, and Commercial Fisheries Development and Management Divisions' staff of the Alaska Department of Fish and Game. The project leaders and their assistants are not fully funded by this project and are supported with general funds from the State of Alaska. These studies are integrated with ongoing studies by the Commercial Fisheries Management and Development Division. Consequently, the EVOS investigations have been integrated into the normal operations of these Divisions for efficiency in completing the objectives of these studies.

7. Contracts:

A sole source contract is proposed (value not to exceed \$50,000) to be awarded to *BioSonics, Inc.* for continuing work in UCI with hydroacoustic equipment. *BioSonics, Inc.* was awarded the original contract in 1992 and 1993 through competitive bid procedures. The experience gained and the recent purchase of *BioSonics, Inc.* equipment for this project make them the logical contractor for the continuation of these studies.

We also propose to develop additional DNA-level stock identification techniques through

the use of contractors. Approaches under consideration include screening microsatellite markers (Bentzen and Wright 1993), sequencing of nuclear genes (Devlin 1993), and screening other regions of mtDNA (Cronin et al. 1993). A single contract or several smaller contracts are proposed (total value less than \$20,000) to be awarded to the most qualified bidder(s) for developmental work on DNA markers for stock identification.

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D. SCHEDULES

Laboratory analyses of 1993 baseline & fishery samples	July 1993 - Apr 1994
Prepare, advertise, and award contracts for 1994 UCI hydroacoustic survey and DNA analysis	Jan - May 1994
Fishery GSI sample collection	June - July 1994
Hydroacoustic assessment	July 1994
Baseline sample collection	June - Sept 1994
Lab analyses of mixtures; modeling of 1994 fishery samples	May - Sept 1994
Final report from hydroacoustic survey contractor	Feb 1995
Draft status report for FY 94	March 1995
Final status report for FY 94	June 1995
Baseline analyses, inseason analyses, annual report	Oct 1994-Sept 1995
Baseline analyses, inseason analyses, final report	Oct 1995-Sept 1996

E. EXISTING AGENCY PROGRAM

For FY 94 the Division of Commercial Fisheries Management and Development (excluding programs in Sport Fish and Subsistence Divisions of ADF&G) will have the following programs: Area management (\$238 K), Research to include catch sampling, escapement monitoring, and offshore test fishing (\$480 K), Biometric, technical, and regional support (\$250 K), and the statewide genetics laboratory in Anchorage (\$500 K).

F. ENVIRONMENTAL COMPLIANCE/PERMIT/COORDINATION STATUS

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. This project received a categorical exclusion under the National Environmental Policies Act (NEPA).

G. PERFORMANCE MONITORING

The performance monitoring of this project is conducted through the checks and balances of the State of Alaska Accounting System within the Commercial Fisheries Management and Development, Habitat and Restoration, and Administration Divisions of the Department of Fish and Game and the Department of Administration. Contractual compliance, personnel hiring, EEO compliance, and other administrative provisions are within the State of Alaska hiring and administrative chains of command and covered in standard operating procedures and administrative regulations. Replacement personnel are readily available by reassignment from permanent and seasonal staff within the Commercial Fisheries Management and Development Division of the ADF&G in Soldotna and Anchorage, when temporary problems are encountered. Filling new positions follows state hiring guidelines when permanent vacancies occur. Project time frames for reports and analysis are maintained through proper planning and integration of these activities within the existing administrative structure of the Commercial Fisheries Management and Development Division.

The scientific and technical aspects of the study are subject to internal review within the Commercial Fisheries Management and Development Division. Publications are submitted through an internal peer review process with the major findings submitted to peer review journals. Reports, work plans, and study design are subject to the peer review process established by the EVOS Board of Trustees and Chief Scientist office. Last year technical aspects of the overescapement studies' findings to date and future plans were reviewed by a panel of international sockeye salmon researchers in a special half day session of the Kokanee and Sockeye Salmon workshop sponsored by the Northern Pacific International Chapter of the American Fisheries Society at Vancouver, B.C. in March 1993. These studies provided the basis for the management programs being developed under this restoration project. Interim annual status reports will be generated with publications being provided in peer review journals and scientific symposia, as significant findings are obtained. The final report will be issued upon completion of the final year of field data collection.

H. COORDINATION OF INTEGRATED RESEARCH EFFORT

The investigations of Kenai River sockeye salmon have been integrated with long term research efforts by the Alaska Department of Fish and Game. These efforts are adult salmon enumeration by hydroacoustic techniques in various river systems, catch and escapement sampling of salmon for age, length, and weight, test fishing at the Central District southern boundary, and juvenile salmon rearing studies. Development of restoration strategies on the Kenai Peninsula are through a review process with the Regional Planning Team and with ADF&G review teams composed of personnel from all ADF&G divisions.

I. PUBLIC PROCESS

The investigations have been and will continue to be part of the public process. Reports have been prepared to the Alaska Board of Fisheries, the Upper Cook Inlet Regional Planning Team, various fisherman's organizations and civic organizations, in addition to the Trustee Council Public Advisory Group process. Scientific and technical review have included the

peer review system established by the Trustee Council. In addition, technical aspects of the studies findings to date and future plans have been reviewed by ADF&G personnel.

J. PERSONNEL QUALIFICATIONS

Kenneth E. Tarbox

Alaska Department of Fish and Game

Commercial Fisheries Management and Development Division

34828 Kalifornsky Beach Road, Suite B

Soldotna, Alaska 99669

(907) 262-9368

EMPLOYMENT:

May, 1980 to Present. Upper Cook Inlet Research Project Leader, Alaska Department of Fish and Game, Soldotna, Alaska. Responsibilities include planning, implementing, supervision, and reporting on various salmon related research and management projects. These involve hydroacoustic enumeration of salmon in glacial systems, defining salmon migratory behavior in both salt and fresh water, evaluation of potential impacts of resource development on habitat and populations, management of the UCI commercial salmon fisheries, stock identification studies using scale or genetic markers, and life history studies of sockeye salmon.

March, 1972 to May, 1980. Project manager and Senior Biologist, Woodward Clyde Consultants, Anchorage, Alaska. Responsibilities included supervision and research for a number of projects. These included an evaluation of existing methodologies for determining instream flow requirements for Alaskan fishes, determining the biological impact of a dredging projects located in lower New York Harbor and Lake Michigan, fishery investigations in the Zayandeh River, Iran, impact assessment of various oil related projects in Virginia, North Carolina, Texas, and Prudhoe Bay, Alaska, and studies and evaluation of impacts associated with nuclear power plants in New Jersey, Louisiana, Indiana, and Pennsylvania.

July, 1970 to March, 1972. Research Assistant, Louisiana Co-operative Fishery Unit, Louisiana State University, Baton Rouge, La. Responsibilities included the design and conduct of a one year investigation of juvenile fish behavior in an estuarine environment.

EDUCATION:

M.S. in Fisheries, 1974. Louisiana State University, Baton Rouge, La.

B.S. in Fisheries Science. 1970. University of Washington, Seattle, Wa.

CERTIFICATIONS:

Fisheries Scientist, Certificate 1165, American Fisheries Society, 1976.

PUBLICATIONS:

Available on request

James E. Seeb, Principal Geneticist

Commercial Fisheries Management and Development
Alaska Department of Fish and Game
Anchorage, Alaska 99518 (907) 267-2385

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, University of Alaska Fairbanks
1988-1990 Assistant Professor, Southern Illinois University
1987-1988 Research Assistant Professor, University of Idaho
1982-1986 Graduate Research Assistant, University of Washington
1980-1982 Fish Biologist, Pacific Fisheries Research, Olympia, WA
1978-1980 Fish Biologist, Washington Department of Fisheries

SELECTED PUBLICATIONS:

- Seeb, J.E., L.W. Seeb, and F.M. Utter. 1986. Use of genetic marks to assess stock dynamics and management programs for chum salmon. *Trans. Amer. Fish. Soc.* 115:448-454.
- Seeb, J.E., and L.W. Seeb. 1986. Gene mapping of isozyme loci in chum salmon (*Oncorhynchus keta*). *J. Hered.* 77:399-402.
- Seeb, J.E., L.W. Seeb, D.W. Oates, and F.M. Utter. 1987. Genetic variation and postglacial dispersal of populations of northern pike (*Esox lucius*) in North America. *Can. J. Fish. Aquat. Sci.* 44:556-561.
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- Gharrett, A. J. B. Riddell, J. Seeb, and J. Helle. 1993. Status of the Genetic Resources of Pacific Rim Salmon. In: J. Cloud, Editor. *Genetic Conservation of Salmonid Fishes*. Plenum Press, New York. pp. 286-292.
- Utter, F. M., J. E. Seeb, and L. W. Seeb. 1993. Complementary uses of ecological and biochemical genetic data in identifying and conserving salmon populations. *Fisheries Research*. *Fish. Res.* 18:59-76.

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

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, University of Alaska Fairbanks
1988-1990 Assistant Professor, Southern Illinois University
1984-1988 Research Assist. Prof., University of Idaho
1978-1981 Fish Geneticist, Pacific Fish. Research, Olympia WA
1977-1979 Geneticist, National Marine Fisheries Service, Seattle

SELECTED PUBLICATIONS:

- Wishard, L. N., J. E. Seeb, F. M. Utter, and D. Stefan. 1984. A genetic investigation of suspected redband trout populations. *Copeia* 1984(1):120-132.
- Seeb, J. E., L. W. Seeb, and F. M. Utter, 1986. Use of genetic marks to assess stock dynamics and management programs for chum salmon. *Trans. Amer. Fish. Soc.* 115:448-454
- Seeb, L. W. and D. R. Gunderson. 1988. Genetic variation and population structure of Pacific ocean perch (*Sebastes alutus*). *Can. J. Fish. Aquat. Sci.* 45:78-88.
- Seeb, L. W., J. E. Seeb, R. L. Allen and W. K. Hershberger. 1990. Evaluation of adult returns of genetically marked chum salmon, with suggested future applications. *American Fisheries Society Symposium* 7:418-425
- Seeb, L. W., J. E. Seeb and A. J. Gharrett. 1990. Genetic marking of fish populations. pp 223-239 in D. H. Whitmore, ed. *Electrophoretic and isoelectric focusing techniques in fisheries management*. CRC Press, Boca Raton, FL.
- Seeb, L. W., J. E. Seeb and J. J. Polovina. 1990. Genetic variation in highly exploited spiny lobster *Panulirus marginatus* populations from the Hawaiian Archipelago. *Fishery Bulletin* 88:713-718.
- Seeb, L. W. and A. W. Kendall. 1991. Allozyme polymorphisms permit the identification of larval and juvenile rockfishes of the genus *Sebastes*. *Environmental Biology of Fishes* 30:191-201.
- Utter, F. M., J. E. Seeb, and L. W. Seeb. 1993. Complementary uses of ecological and biochemical genetic data in identifying and conserving salmon populations. *Fisheries Research. Fish. Res.* 18:59-76.

K. BUDGET

See attached.

Table 1. Enzymes or proteins screened in Cook Inlet sockeye salmon. Enzyme nomenclature follows Shaklee et al. (1990), and locus abbreviations are given.

Enzyme or Protein	Enzyme Number	Locus	Tissue
Aspartate aminotransferase	2.6.1.1	<i>sAAT-1,2</i>	Heart
		<i>sAAT-3</i>	Eye
		<i>sAAT-4</i>	Liver
		<i>mAAT-1</i>	Heart
		<i>mAAT-2</i>	Liver
		<i>mAAT-3</i>	Liver
Acid phosphatase	3.1.3.2	<i>ACP-1</i>	Liver
Adenosine deaminase	3.5.4.4	<i>ADA-1</i>	Muscle
		<i>ADA-2</i>	Heart
Aconitate hydratase	4.2.1.3	<i>MAH-1,2</i>	Heart
		<i>MAH-3</i>	Heart
		<i>MAH-4</i>	Heart
		<i>SAH</i>	Liver
Adenylate kinase	2.7.4.3	<i>AK</i>	Eye
Alanine aminotransferase	2.6.1.2	<i>ALAT</i>	Muscle
Creatine kinase	2.7.3.2	<i>CK-A1</i>	Muscle
		<i>CK-A2</i>	Muscle
		<i>CK-B</i>	Eye
		<i>CK-C1</i>	Eye
		<i>CK-C2</i>	Eye
Esterase-D	3.1.1.-	<i>ESTD</i>	Muscle
Fructose-biphosphate aldolase	4.1.2.13	<i>FBALD-3</i>	Eye
		<i>FBALD-4</i>	Eye
Formaldehyde dehydrogenase (Hydroxyacylglutathione hydrolase (HAGH))	1.2.1.46	<i>FDH</i>	Liver

Table 1. Continued.

Enzyme or Protein	Enzyme Number	Locus	Tissue
Fumarate hydratase	4.2.1.2	<i>FH</i>	Muscle
β -N-Acetylgalactosaminidase	3.2.53	<i>\beta</i> GALA	Liver
Glyceraldehyde-3-phosphate dehydrogenase	1.2.1.12	<i>GAPDH-1</i>	Muscle
		<i>GAPDH-2</i>	Heart
		<i>GAPDH-3</i>	Heart
		<i>GAPDH-4</i>	Eye
		<i>GAPDH-5</i>	Eye
Guanine deaminase	3.5.4.3	<i>GDA-1</i>	Liver
		<i>GDA-2</i>	Liver
N-Acetyl- β -glucosaminidase	3.2.1.53	<i>\beta</i> GLUA	Liver
Glycerol-3-phosphate dehydrogenase	1.1.1.8	<i>G3PDH-1,2</i>	Muscle
		<i>G3PDH-3</i>	Heart
		<i>G3PDH-4</i>	Heart
Glucose-6-phosphate isomerase	5.3.19	<i>GPI-B1,2</i>	Muscle
		<i>GPI-A</i>	Muscle
Glutathione reductase	1.6.4.2	<i>GR</i>	Eye
Isocitrate dehydrogenase (NADP +)	1.1.1.42	<i>mIDHP-1</i>	Heart
		<i>mIDHP-2</i>	Heart
		<i>sIDHP-1</i>	Liver
		<i>sIDHP-2</i>	Liver
L-Lactate dehydrogenase	1.1.1.27	<i>LDH-A1</i>	Muscle
		<i>LDH-A2</i>	Muscle
		<i>LDH-B1</i>	Eye
		<i>LDH-B2</i>	Liver

Table 1. Continued.

Enzyme or Protein	Enzyme Number	Locus	Tissue
		<i>LDH-C</i>	Eye
α Mannosidase	3.2.1.24	<i>αMAN</i>	Liver
Malate dehydrogenase	1.1.1.37	<i>sMDH-A1,2</i>	Heart
		<i>sMDH-B1,2</i>	Heart
		<i>mMDH-1</i>	Heart
		<i>mMDH-2</i>	Muscle
		<i>mMDH-3</i>	Muscle
Malic enzyme (NADP +)	1.1.1.40	<i>sMEP-1</i>	Muscle
		<i>sMEP-2</i>	Liver
		<i>mMEP</i>	Muscle
Mannose-6-phosphate isomerase	5.3.1.8	<i>MPI</i>	Liver
Dipeptidase	3.4.-.-	<i>PEPA</i>	Eye
Tripeptide aminopeptidase	3.4.-.-	<i>PEPB-1</i>	Heart
Peptidase-C	3.4.-.-	<i>PEPC</i>	Eye
Proline dipeptidase	3.4.13.9	<i>PEPD-1</i>	Heart
Peptidase-LT	3.4.-.-	<i>PEPLT</i>	Muscle
Phosphogluconate dehydrogenase	1.1.1.44	<i>PGDH</i>	Liver
Phosphoglucomutase	5.4.2.2	<i>PGM-1</i>	Heart
		<i>PGM-2</i>	Muscle
Phosphoglycerate kinase	2.7.2.3	<i>PGK-1</i>	Eye
		<i>PGK-2</i>	Eye
Pyruvate kinase	2.7.1.40	<i>PK-1</i>	Heart
		<i>PK-2</i>	Heart
Purine-nucleoside phosphorylase	2.4.2.1	<i>PNP-1</i>	Eye
Superoxide dismutase	1.15.1.1	<i>sSOD-1</i>	Liver

Table 1. Continued.

Enzyme or Protein	Enzyme Number	Locus	Tissue
Triose-phosphate isomerase	5.3.1.1	<i>mSOD</i>	Heart
		<i>TPI-1,2</i>	Eye
		<i>TPI-3</i>	Eye
		<i>TPI-4</i>	Eye

Table 2. Restriction enzymes to be used in DNA pilot study.

Restriction Enzyme		Recognition Site
1	<i>Apa I</i>	GGGCC'C
2	<i>BamH I</i>	G'GATCC
3	<i>Bcl I</i>	T'GATCA
4	<i>Bgl I</i>	GGCN ₄ 'NGGC
5	<i>Bgl II</i>	A'GATCT
6	<i>BstE II</i>	G'GTNACC
7	<i>BstU I</i>	CG'CG
8	<i>Dpn II</i>	'GATC
9	<i>EcoR I</i>	G'AATTC
10	<i>EcoR V</i>	GAT'ATC
11	<i>Hae III</i>	GG'CC
12	<i>Hha I</i>	GCG'C
13	<i>Hind III</i>	A'AGCTT
14	<i>Hinf I</i>	G'ANTC
15	<i>Kpn I</i>	GGTAC'C
16	<i>Mse I</i>	T'TAA
17	<i>Msp I</i>	C'CGG
18	<i>Nci I</i>	CC'(G/C)GG
19	<i>Pst I</i>	CTGCA'G
20	<i>Sac I</i>	GAGCT'C
21	<i>Sac II</i>	CCGC'GG
22	<i>Sau96 I</i>	G'GNCC
23	<i>Sca I</i>	AGT'ACT
24	<i>Stu I</i>	AGG'CCT
25	<i>Taq I</i>	T'CGA
26	<i>Xba I</i>	T'CTAGA
27	<i>Xho I</i>	C'TCGAG

APPENDIX A

25 May 1993

Collection of Finfish Genetic Samples

ADF&G Genetics Laboratory, Anchorage

I. General info

We use tissue samples from muscle, liver, heart, and eye from individual fish to determine the genetic characteristics of a particular run or stock of fish. The most important thing to remember in collecting samples is that tissues need to be as fresh and as cold as possible at all times.

II. Sample size

A sample size of 50-100 adult fish is preferred for the baseline electrophoretic study. Samples of juveniles are statistically less desirable and sample sizes will need to be larger than for adults; generally a sample size of 150-200 juveniles is necessary.

III. Tissue sampling

A. General set up

We use four tissues (muscle, liver, eye, and heart) for protein electrophoresis. Working fast is necessary, so it is best to try to get set up in as comfortable a place as possible. You might use a portable table, piece of plywood, or anything to give you a surface at a good height. Before sampling (night before?), label tubes using lab markers or adhesive labels (provided in sampling kit). Place the prepared tubes in the racks provided. Four separate tubes, corresponding to the four tissues, should be labeled for each individual. The following code should be used:

Species code * see instructions for each project

Location code * see instructions for each project

Individual # # i.e. 01, 02, 03....100

Tissue M,L,E,H (muscle, liver, eye, heart)

B. Use of liquid nitrogen

We will be using a liquid nitrogen container to immediately freeze the tissues. Inside the liquid nitrogen container are 6 cylindrical canisters. We

have shipped special test tubes called "cryotubes" in which to place the samples. These cryotubes have plastic seals and screw on caps to withstand liquid nitrogen storage. Five to six tubes are stored in a cane.

The working time of the liquid nitrogen container under normal conditions is 81 days (35VHC) or 50 days (18HC). To prolong the liquid nitrogen, samples can be pre-frozen (if a freezer or dry ice is available) and added in a group to minimize the number of times the container is opened. The liquid nitrogen level can be checked periodically with a flashlight or actually measured with a stick (2.3 liters/inch in 35VHC; 1.25 liters/inch in 18HC).

"Large" 35 VHC container:

30 canes will fit in each of the six canisters. 5 cryovials will fit on a cane comfortably or 6 in a pinch. Total capacity is 900 - 1080 tubes.

"Small" 18HC container:

17 canes will fit in each of the six canisters. 5 to 6 cryovials will fit on a cane. The total capacity is 510 - 612 Nalgene tubes.

Safety with liquid nitrogen:

1. Wear gloves, protective eyewear, and protective footwear when placing samples in container. Liquid nitrogen boils at -196° , and it will spit and boil when samples are added.
2. Do not tip the tank over as it does not seal.
3. Keep lid on liquid nitrogen container at all times when you are not placing samples in it.
4. Use a small cooler with ice, snow, or blue ice to hold canes until an adequate number are collected to be put in liquid nitrogen container. Depending on the conditions and the speed of sampling, place samples in liquid nitrogen within about one hour of sampling.
5. Use liquid nitrogen only in well ventilated areas (usually not a problem in the field). Avoid directly breathing the vapor.
6. Hazardous Materials Forms need to be filled out when shipping a filled liquid nitrogen container by air cargo.

B. Actual sampling

Please take samples from freshly killed fish. We find it easiest to set up four canes simultaneously and organize the samples in canes by tissue. Thus, muscle tissue from fish 1-5 would all be in one cane.

Fill the tubes approximately 3/4 full or to the 1.8 ml mark, leaving air space at the top. Overfilling the tubes can cause them to burst when frozen. Please minimize the amount of blood, dirt, skin, and fat in the sample.

1. Muscle

Muscle samples should be "white" muscle, not muscle from along the lateral line. Use a piece of muscle dorsal to the lateral line. If you have trouble getting the tissue into the tubes, cut it into smaller pieces.

2. Liver

The liver is (generally) located on the fish's left side, just behind the pectoral fin. An L-shaped incision slicing down ventrally behind the pectoral fin then caudally along the belly works well. Please do not include the gall bladder (the small green/yellow sac of fluid attached to the liver).

3. Heart

Once you have taken the liver, it is easy to get the heart by just opening the belly incision towards the head.

4. Eye

There are two ways to take the eyes. If the eyes are small enough (juveniles), they can be placed intact into a cryotube. This is the easiest method. If they are too large, you must pipette out the liquid and black retinal fluid. Using a sharp scalpel, cut a small slit in the surface of the eye, then insert a pipette into the slit and suck out the fluid and black retinal material. Squirt this into the cryotube.

We appreciate your help with the sampling. If you have any questions, please give us a call.

Laboratory (Lori Wagoner)	267-2454
Chris Habicht	267-2169
Lisa Seeb	267-2249

APPENDIX B

Guidelines for GSI Fishery Sampling

I. General info

We use tissue samples from muscle, liver, heart, and eye from individual fish to determine the genetic characteristics and profile of the fishery. Fish sampled from the fishery will be of varying quality. However, once you begin sampling, the tissues need to be handled carefully and kept as cold as possible at all times.

II. Sample size and design

A sample size of 400 fish per sampling period per area is needed. Our goal is to obtain as dispersed and randomized sampling of the fishery as possible. If feasible, sample in 100 fish groups to spread sampling across the sampling period. If this is not possible, 400 fish can be set aside and sampled as a group.

III. Tissue sampling

A. General set up

We use four tissues (muscle, liver, eye, and heart) for protein electrophoresis. **It is critical in fishery sampling to insure that all tissues from one individual are correctly marked.** We must be able to merge data across tissues for each individual

Working fast is necessary, so it is best to try to get set up in as comfortable a place as possible. You might use a portable table, piece of plywood, or anything to give you a surface at a good height. Before sampling (night before?), label tubes using adhesive labels (provided in sampling kit). Place the prepared tubes in the racks provided. Four separate tubes, corresponding to the four tissues, should be labeled for each individual. The following code should be used:

Species code	* see instructions for each project
Location code	* see instructions for each project

Individual # # i.e. 01, 02, 03....100

Tissue M,L,E,H (muscle, liver, eye, heart)

B. Use of liquid nitrogen

We will be using a liquid nitrogen container to immediately freeze the tissues. Inside the liquid nitrogen container are 6 cylindrical canisters. We have shipped special test tubes called "cryotubes" in which to place the samples. These cryotubes have plastic seals and screw on caps to withstand liquid nitrogen storage. Five to six tubes are stored in a cane.

The working time of the liquid nitrogen container under normal conditions is 81 days (35VHC) or 50 days (18HC). To prolong the liquid nitrogen, samples can be pre-frozen (if a freezer or dry ice is available) and added in a group to minimize the number of times the container is opened. The liquid nitrogen level can be checked periodically with a flashlight or actually measured with a stick (2.3 liters/inch in 35VHC; 1.25 liters/inch in 18HC).

"Large" 35 VHC container:

30 canes will fit in each of the six canisters. 5 cryovials will fit on a cane comfortably or 6 in a pinch. Total capacity is 900 - 1080 tubes.

"Small" 18HC container:

17 canes will fit in each of the six canisters. 5 to 6 cryovials will fit on a cane. The total capacity is 510 - 612 Nalgene tubes.

Safety with liquid nitrogen:

1. Wear gloves, protective eyewear, and protective footwear when placing samples in container. Liquid nitrogen boils at -196° , and it will spit and boil when samples are added.
2. Do not tip the tank over as it does not seal.
3. Keep lid on liquid nitrogen container at all times when you are not placing samples in it.
4. Use a small cooler with ice, snow, or blue ice to hold canes until an adequate number are collected to be put in liquid nitrogen container. Depending on the conditions and the speed of sampling, place samples in liquid nitrogen within about one hour of sampling.
5. Use liquid nitrogen only in well ventilated areas (usually not a problem in the field). Avoid directly breathing the vapor.
6. Hazardous Materials Forms need to be filled out when shipping a filled liquid nitrogen container by air cargo.

B. Actual sampling

We find it easiest to set up four canes simultaneously and organize the

EXXON VALDEZ TRUSTEE COUNCIL
 1994 Federal Fiscal Year Project Budget
 October 1, 1993 - September 30, 1994

Project Description: Kenai River Sockeye Salmon Restoration - This project is a continuation of 93015 and a merger with 93012 (Genetic Stock Identification of Kenai River Sockeye Salmon). Its goal is to restore depressed Kenai River sockeye stocks by directing fisheries away from impacted stocks and allowing fishing on healthy stocks. This objective is accomplished through improved stock identification capabilities using analyses of genetic data and parasite information, and improved stock assessment capabilities using hydroacoustics.

Budget Category:	1993 Project No. 93015 Authorized FFY 93	'93 Report/ '94 Interim* FFY 94	Remaining Cost** FFY 94	Total FFY 94	FFY 95	Comment
Personnel	\$276.4	\$94.7	\$113.3	\$208.0	\$208.0	FFY 95 cost may vary depending upon results from FFY 94.
Travel	\$14.8	\$7.0	\$2.1	\$9.1	\$9.1	
Contractual	\$252.9	\$2.0	\$88.1	\$90.1	\$90.1	
Commodities	\$44.4	\$3.0	\$34.4	\$37.4	\$37.4	
Equipment	\$112.3	\$0.0	\$24.0	\$24.0	\$24.0	
Capital Outlay	\$0.0	\$0.0	\$0.0	\$0.0	\$0.0	
Subtotal	\$700.8	\$106.7	\$261.9	\$368.6	\$368.6	
General Administration	\$59.2	\$14.3	\$23.2	\$37.5	\$37.5	
Project Total	\$760.0	\$121.0	\$285.1	\$406.1	\$406.1	
Full-time Equivalents (FTE)	4.0	1.5	3.0	4.5	4.5	
Dollar amounts are shown in thousands of dollars.						
Budget Year Proposed Personnel: Position Description	Reprt/Intrm Months	Reprt/Intrm Cost	Remaining Months	Remaining Cost		
Reprt Fisheries Biologist III & IV	4.0	\$26.0	0.0	\$0.0		
Fisheries Biologist I & II	5.0	\$24.0	4.0	\$16.8		
Biometrician I & III	4.0	\$27.8	0.0	\$0.0		
Field Office Assistant	2.0	\$7.0	0.0	\$0.0		
Fisheries Tech. III	1.0	\$3.5	3.0	\$9.7		
Fisheries Tech. II	1.0	\$3.0	28.0	\$80.1		
Program Manager	0.5	\$3.4	1.0	\$6.7		
Personnel Total	17.5	\$94.7	36.0	\$113.3		
					NEPA Cost:	\$5.0
					*Oct 1, 1993 - Jan 31, 1994	
					**Feb 1, 1994 - Sep 30, 1994	

07/14/93

1994

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Project Number: 94255
 Project Title: Kenai River Sockeye Salmon Restoration
 Agency: AK Dept. of Fish & Game

**FORM 2A
 PROJECT
 DETAIL**

EXXON VALDEZ TRUSTEE COUNCIL

1994 Federal Fiscal Year Project Budget

October 1, 1993 - September 30, 1994

Travel:		Reprt/Intrm	Remaining
Reprt	4 RT Anchorage/Cordova @ \$350/trip + 1 day per diem @ \$150/day	\$2.0	\$0.0
	Admin. travel- 10 RT Kenai/Anchorage @ \$150/trip	\$1.5	\$0.0
	5 meetings w/ peer reviewers, American Fisheries Society, etc. @ \$350/trip + 1 day per diem @ \$150/day	\$2.5	\$0.0
	Per diem.	\$1.0	\$0.0
	Field travel (2 RT Anchorage/PWS)	\$0.0	\$1.1
	Admin. travel 5 RT Kenai/Anchorage	\$0.0	\$1.0
Travel Total		\$7.0	\$2.1
Contractual:			
Reprt	Freight, postage, etc.	\$0.8	\$0.8
	Air charter for stock identification (25 hours x \$400/hour)	\$0.0	\$10.0
	Vehicle rental (2 vehicles for 6 months @ \$400/month)	\$0.0	\$4.8
	Fuel	\$0.0	\$0.4
	Long distance phone charges	\$0.5	\$1.1
	Computer/other repair	\$0.7	\$1.0
	Contract for Cook Inlet hydroacoustics survey for collection and analysis of field data and preparation of a final report.	\$0.0	\$50.0
	Contract for DNA analysis	\$0.0	\$20.0
Contractual Total		\$2.0	\$88.1

07/14/93

Page 2 of 3

1994

Printed: 3/15/94 11:48 AM

Project Number: 94255
Project Title: Kenai River Sockeye Salmon Restoration
Agency: AK Dept. of Fish & Game

FORM 2B
PROJECT
LAIL

EXXON VALDEZ TRUSTEE COUNCIL
 1994 Federal Fiscal Year Project Budget
 October 1, 1993 - September 30, 1994

Commodities:	Reprt/Intrm	Remaining
Reprt Publishing and computer supplies	\$1.5	\$0.0
Computer software upgrades	\$1.5	\$0.0
Computer supplies for hydroacoustics (cables, charts, connectors)	\$0.0	\$13.4
Computer software upgrade	\$0.0	\$1.0
Biochemicals (tissue preservatives, alcohol, stains, fixatives, etc.)	\$0.0	\$12.0
Lab supplies (tubes, pipettes, gel rigs, plasticware)	\$0.0	\$5.0
Office supplies (paper, pens, pencils, markers, etc.)	\$0.0	\$3.0
Commodities Total	\$3.0	\$34.4
Equipment:		
Network equipment	\$0.0	\$2.0
Computer memory 8 meg upgrade	\$0.0	\$1.0
Sampling - raft, tent replacements	\$0.0	\$2.0
Freezer	\$0.0	\$8.0
Laboratory computer	\$0.0	\$5.0
DC power supplies	\$0.0	\$3.0
Microcentrifuge	\$0.0	\$1.0
Storage equipment	\$0.0	\$2.0
Equipment Total	\$0.0	\$24.0

07/14/93

1994

Page 3 of 3

Printed: 3/15/94 11:48 AM

Project Number: 94255
 Project Title: Kenai River Sockeye Salmon Restoration
 Agency: AK Dept. of Fish & Game

**FORM 2B
 PROJECT
 DETAIL**

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EXXON VALDEZ TRUSTEE COUNCIL
FY 94 DETAILED PROJECT DESCRIPTION

A. COVER PAGE

Project title:	Sockeye Salmon Overescapement
Project ID number:	94258
Project type:	Monitoring
Name of project leader(s):	Dana Schmidt and Ken Tarbox
Lead agency:	ADF&G
Cooperating agencies:	USFWS and NBS
Cost of project/FY 94:	\$850
Cost of project/FY 95:	\$592
Cost of Project/FY 96 and beyond:	\$500
Project Start-up/Completion Dates:	October 1, 1993 / September 30, 1994
Geographic area of project:	Kodiak Island and the Kenai Peninsula
Name of lead agency project manager:	Dr. Joseph R. Sullivan

B. INTRODUCTION

This study is a continuation of the oil spill damage assessment program initiated in 1990 (Schmidt and Tarbox, 1993). The continuing program reflects modifications based on the FY93 study results. Recommendations provided by an international review team of sockeye salmon experts provided at a March 15, 1993 meeting at Vancouver, B.C. have been incorporated.

Commercial fishing for sockeye salmon in 1989 was curtailed in upper Cook Inlet, the outer Chignik districts, and the Kodiak areas due to presence of oil in the fishing areas from the EVOS. As a result, the number of sockeye salmon entering four important sockeye producing systems (Kenai/Skilak, Chignik/Black, Red, and Frazer Lakes) and two less important lake systems (Akalura and Afognak or Litnik lakes) greatly exceeded levels that are thought to be most productive. Sockeye salmon spawn in lake associated river systems. Adult salmon serve an extremely important role in the ecosystem, providing food for marine mammals, terrestrial mammals, and birds. Additionally, carcass decomposition serves to charge fresh water lake systems with important nutrients. Juvenile salmon which rear in lakes for one or two years serve as a food source for a variety of fish and mammals. Sockeye salmon are also an important subsistence, sport, and commercial species. The ex-vessel value of the commercial catch of sockeye from these lake systems has averaged about \$42 million per year since 1979, with the 1988 catch worth \$115 million. Sockeye salmon returns to the Kenai River system support some of the largest recreational fisheries in the State.

Overly large spawning escapements may result in poor returns by producing more rearing juvenile sockeye than can be supported by the nursery lake's productivity (Kyle et al. 1988). In general, when rearing fish abundance greatly exceeds the lake's carrying capacity, prey resources are altered by changes in species and size composition (Mills and Schiavone 1982, Koenings and Burkett 1987, Kyle et al. 1988) with concomitant effects on all trophic levels (Carpenter et al. 1985). Because of such changes, juvenile sockeye growth is reduced, mortality increases, larger percentages holdover for another year of rearing; and the poor quality of smolts increases marine mortality. Where escapements are two to three times normal levels, the resulting high juvenile densities crop the prey resources to the extent that more than one year is required to return to normal productivity. Rearing juveniles from subsequent brood-years suffer from both the poor quality of forage and from the increased competition for food by holdover juveniles (Townsend 1989; Koenings and Kyle 1991). This is the brood-year interaction underlying cyclic variation in the year class strength of anadromous fish.

This project continues examining the effects of large 1989 spawning escapements on the resulting progeny and associated foraging habitat for a select subset of the above mentioned sockeye nursery lakes. Three impacted lake systems where the 1989 escapements were more than twice the desired levels (Kenai/Skilak in Upper CI; Red and Akalura lakes on Kodiak Island) were selected. Upper Station Lake which is near the two impacted lakes on Kodiak did not receive a large escapement and has been examined as a control. Because of significant differences in the

basin morphology and limnology, Upper Station has proved relatively ineffective as a control (Schmidt and Tarbox 1993). Beginning in 1994, Frazer Lake will be used for future comparisons. Because this lake has undergone detailed study in the past (Kyle et al. 1988) and has continued funding from other sources, minimal funding is necessary to provide for data collection to insure comparisons with Akalura and Red lakes. Frazer has significant differences from Red and Akalura lakes as Frazer Lake was a natural barriered system with the sockeye salmon run established by introductions and maintained by fishway passage within the past three decades. This lake has been fertilized in the late 1980's as well, unlike the other lakes. Similarly, Tustumena Lake on the Kenai Peninsula received normal escapements and is used as a control for the Kenai River systems. This lake differs primarily in the increased natural turbidity levels and a history of modest stocking of sockeye salmon fry.

Schmidt and Tarbox (1993) report the results through 1992 on these ongoing investigations. In addition, the study proposal reflects results of data collected in the spring of 1993. These studies suggest continued poor smolt production from the Kenai River, despite normal smolt production from Tustumena Lake, the control for this system. The 1992 and 1993 (preliminary) data indicate Red Lake zooplankton communities and nutrient levels have recovered to the level measured in 1986, prior to the oil spill (Schmidt and Tarbox, 1993) and will be discontinued in 1994. Smolt numbers appear to be lagging but adult forecasts for returns in 1994 suggest escapement goals will be met; therefore management actions will be used as the primary method for restoration. Smolt numbers will be used to forecast future returns and provide assistance to managers in future harvest management decisions. Akalura Lake demonstrated poor zooplankton densities with low smolt numbers. The 1994 adult run is not expected to meet escapement requirements. A restoration plan for Akalura lake will be produced during the winter of 1993-94 based on analysis of data collected to date.

The 1993 smolting information from the Kenai system lakes indicate poor smolt production. In addition, the Russian River smolt project indicated very poor production, suggesting this system may also be below production expectations. This system was first identified as having suspect production when the 1992 smolt count from the lower Kenai failed to produce sufficient numbers of age 2 smolt to account for historic Russian River smolt migrations. This project suggests further investigations into the Russian River are warranted.

C. PROJECT DESCRIPTION

1. Resources and/or Associated Services:

The studies are located on Kodiak Island and the Kenai Peninsula. Recent findings (Schmidt and Tarbox, 1993) have suggested major economic damage to commercial, subsistence, and sport fisheries may result because of the over-

escapement event associated with the fisheries closures on the Kenai River sockeye salmon stocks caused by the 1989 oil spill. Smolt numbers emigrating from the Kenai River in the spring of 1992 and 1993 were less than one-fiftieth the numbers estimated in 1989. This suggests a likely possibility of future returns below existing escapement goals. Red River smolt numbers from the 1989 escapement on Kodiak Island are estimated to return at rates which will provide for minimal commercial harvests if average marine survival occurs.

In addition to monitoring the damage extent, the mechanism that lead to the collapse requires definition. These studies essentially follow the pattern established in the original 1990 study plan but with significant modifications to accommodate recent findings.

2. Relation to Other Damage Assessment/Restoration Work:

Restoration studies funded by the trustees that are based on the results of these studies include 94255 and 94504. These restoration projects provide for improved management of Cook Inlet sockeye salmon runs, anticipated to return in reduced numbers because of the failure of the Kenai River system sockeye salmon.

3. Objectives:

The following objectives are altered based on input from peer reviewers of the 1992 progress report and proposed revisions to the 1994 study program..

- a. Estimate critical biological attributes (number, age, size) of both resident and migrant juvenile sockeye in overescaped and normal escaped sockeye salmon nursery lakes of the Kenai Peninsula and Kodiak Island.
- b. Determine effects on smolt production and subsequent adult returns caused by large escapements resulting from fishery closures after the EVOS. These effects will be inferred by studying the changes in the rearing capacity of selected nursery lakes which were either affected or unaffected by the oil spill. Data used for these inferences include:
 - (1). age and growth of juveniles and smolts
 - (2). nursery area nutrient budgets and plankton populations.
 - (3). seasonal, diel, and vertical distribution of zooplankton species which are the known prey of sockeye salmon in Skilak, Kenai, and Tustumena Lake; and
 - (4). seasonally available zooplankton biomass in these lakes and

the relationship of this biomass to ambient temperature and light.

- c. Develop a pilot research project to determine experimentally the cause of the decline and potential restorative actions.

ADF&G in cooperation with the regional research staff of the U.S. Fish and Wildlife Service and the Refuge staff of the Kenai National Wildlife Refuge will develop a pilot research project to further define the mechanism of sockeye salmon decline and determine the feasibility of alternative restoration opportunities.

4. Methods:

Numbers of adult sockeye salmon that entered selected spawning systems outside PWS prior to and during 1989 have been estimated at weir stations or by sonar. This information was collected during projects routinely conducted by the ADF&G as part of their resource management program. Optimal escapement levels, which on the average should produce maximum sustained yield, have been based on either past relationships between spawners and returning progeny or the extent of available spawning and rearing habitat. The baseline program will continue at each site including but not limited to estimates of adult sockeye escapement and collection of scales for age analysis.

For each of the lake systems identified, the response (abundance, growth, and freshwater age) of rearing juveniles will be studied. Because of the significance and magnitude of the findings on Red Lake, and on Skilak/Kenai lakes, these studies will continue until observed effects on growth and the limnetic community of the lake ecosystems recovers to pre-spill conditions.

The timeline of the 1994 studies is outlined on Table 1. This table depicts the sampling schedule for the integrated limnological studies and fisheries studies on the Kenai Peninsula. The total number of juvenile sockeye in the Kenai Peninsula lakes will be estimated through hydroacoustic surveys conducted during all years up until recovery of the system is observed. Age and size information will be obtained from samples of juvenile sockeye collected from concurrent mid-water trawl netting surveys. Survey transect designs for hydroacoustic sampling and tow-netting have been established for Kenai and Skilak lakes (Tarbox and King 1989) and Tustumena Lake (Kyle 1992). Depending on densities of rearing juvenile sockeye salmon, estimates of fish densities will be made for each transect either by echo integration or by echo counting. Total fish population estimates will be computed, by summing transect populations, along with 95% confidence intervals (Kyle 1989). Additional studies of the vertical distribution of Skilak Lake sockeye will be conducted simultaneously with population estimates with an additional sampling period, for vertical sampling only, in November, 1993.

Freshwater growth and age of sockeye salmon rearing juveniles from all study systems will be determined from scale and possibly otolith measurements made either by direct visual analysis of scales or using an Optical Pattern Recognition system. In cases where data are available (e.g., Kenai and Skilak Lakes and Tustumena Lake), growth of progeny from the 1989 spawning escapements will be compared with growth or size of progeny during prior years.

The total number of smolt migrating from each system will be estimated with a mark-recapture study using inclined plane traps after Kyle (1983), and King et al. (1991). Smolt will be captured in traps, sampled for age and size information, marked with Bismark Brown Y (a biological dye), and transported upstream of the traps and released for subsequent recapture (Rawson 1984). Periodic retesting will determine the capture efficiency of the traps under changing river conditions during the spring. Total population estimates (with 95% confidence intervals) will be made using catch efficiencies, and weekly number weighted smolt size and age information will be calculated using a computer spreadsheet developed by Rawson (personal communication, 1985). Smolt programs consistent with those for the study lakes are continuing for Tustumena Lake (Kyle 1992).

On the Kenai River, the smolt operation will require expansion to include the Russian River. This lake system apparently now is the dominant producer of sockeye salmon smolt and is upriver from the current smolt project on the mainstem Kenai River. To determine the production of smolt from the Kenai River mainstem, estimates of smolt production from the Russian River lake system must be completed to separate normal Russian River production from the smolt production of sockeye salmon rearing in Skilak and Kenai lakes. These methods are being established to insure current projections of smolt production from the Kenai River lake systems are not an artifact of some unknown sampling bias.

In 1993 we fished one trap in the Russian River to estimate the smolt migration. We caught few fish (less than 5000) and the catch rate was 5%. In 1994, increased trap catch rates will be required to insure we are not missing fish in this clear water system.

The current length frequency data on sockeye smolts at the Kenai mainstem river smolt traps indicates that we may be missing a portion of the Russian River smolt outmigration. With the excellent genetic separation of Russian River sockeye from mainstem Kenai fish we should be able to separate these two components in the catch. This will allow for a better total smolt estimate and verification of the Russian River smolt trap data.

Because the smolt data for 1993 indicated that the Russian River lake systems may be experiencing similar declines in production as the mainstem Kenai River. The 1989 escapement into this system was 138,000 adults, which is far in excess of the minimum 30,000 goal. Therefore, to evaluate the current production potential and impacts of large escapements, limnological and fry hydroacoustic/tow net surveys of the Russian lakes are proposed. Techniques duplicate those used on other systems.

In the two Kenai Peninsula lakes, early spring and late fall sampling of fry will be conducted. The reason for the additional sampling period is that approximately 50% of the weight gain from fry to smolt in the Kenai River system occurs outside of the current sampling regime. If poor survival is occurring because of limitations in rearing habitat quality during this period, these data are crucial for determining the validity of fry density causing decreased over-wintering survival. Based on peer review comments, hydro-acoustic studies of fry abundance will be conducted into the fall, 1994 to track and sample the juvenile fish until cold weather prevents further studies. This is based on the assumption that most of the density dependent mortality occurs in early winter (peer review comments, Hyatt and Hilborn).

Studies on Kodiak Island will be reduced because of recent findings. These include elimination of the smolt weir counts on Red River; relying on mark/recapture studies with smolt traps will be used to estimate smolt abundance in 1994. In 1992 the hydro-acoustic surveys were eliminated on these lakes because of interference of stickleback with the population estimates. Samples of fall fry for age, weight, and length will continue to be collected. The variation and differences in Upper Station lakes suggest this system is inadequate as a control for Red Lake and Akalura Lake. Therefore, Frazer lake will be used as a control in the future. Monitoring of this system is primarily conducted by general fund expenditures of the Alaska Department of Fish and Game. A minor modification will be made to the program to insure compatibility with the monitoring continuing on Akalura and Red lakes. Funding from these studies will be used to augment the regular smolt monitoring program. A second inclined plane trap will be used to ensure that adequate samples are obtained for more accurately describing smolt population numbers and AWL characteristics to insure similar precision with the Red and Akalura lakes studies. The continued poor smolt production in the spring of 1993 (600,000 Red Lake, 90,000 Akalura Lake) suggest continued monitoring of these systems is warranted.

Limnological data will be collected to monitor the response of the lakes to high juvenile rearing densities and their recovery once escapement levels decline. Table 1 provides a time-line of these studies with and reflects the integration with the fisheries investigations previously discussed. These data will be used to estimate carrying capacity parameters of euphotic volume, nutrient budgets (carcass enrichment), and zooplankton biomass, body-sizes, and composition shifts. Approximately six limnology surveys will be conducted at two or more stations, to determine zooplankton species abundance and body-sizes, nutrient chemistry, and phytoplankton abundance for Kenai/Skilak, Tustumena, Red, and Frazer lakes. Methods for limnological studies are detailed in Koenings et al. (1987).

In cases where seasonal data are available (i.e. Kenai and Skilak lakes), limnological parameters taken during residence of the juveniles from the 1989 spawning escapements will be compared to parameters within these systems during prior years.

Although in the Kenai River system smolt enumerations and fall fry estimates during 1991, 1992 and the spring of 1993 produced very low numbers, zooplankton biomass estimates in Skilak Lake, the major sockeye salmon producer, has not undergone similar levels of decline. To further understand the mechanism that may regulate the survival of sockeye salmon juveniles in this lake, early spring tow netting for juvenile salmon was conducted. Failure to collect adequate numbers prompted limited distribution studies of juvenile sockeye in the lakes by use of sonar. These data indicated concentrations during the day near 40 meters but in very low abundances (Schmidt and Tarbox, 1993). These findings prompted limited vertical sampling of the zooplankton community to determine depth distribution. During the day, most of the zooplankton biomass was concentrated at the same depth as the fish with increased surface concentrations during the night. Since light extinction during the spring occurred near 15 meters in this lake and the lake was isothermal at 2.5° C, this distribution pattern seemed peculiar. Since sockeye salmon are principally sight feeders, this indicated that much of the biomass was unavailable for feeding. The control lake, Tustumena, indicated that the same species of zooplankton (*Diaptomus*) did not exhibit a similar vertical distribution. Pearre(1979) and Enright(1977) discussed possible causative mechanisms of various patterns of vertical distribution. One possible mechanism that would explain the difference is food satiation. By having heavy cropping of the zooplankton community, the zooplankton respond by no longer competing for limited food resources and are able to sustain sufficient nutrition with relative minor amounts of time at depths that produce phytoplankton. At these depths they are also susceptible to sight feeding predators (sockeye salmon). Thus, although the high turbidity and cold temperatures of Tustumena produce more limited biomass of zooplankton, their continual presence in the surface light layer makes them much more vulnerable to feeding sockeye. We are also examining if the egg bearing component of the population of zooplankton is being cropped at higher rates which in turn may cause a loss of needed lipids for overwintering survival of sockeye juveniles.

To test these hypotheses, much more intensive sampling of the diel and seasonal distribution of plankton in the glacial lakes of the Kenai Peninsula was initiated in the summer of 1993 by use of a towable optical zooplankton counter(OPC). Because of the limited number of species and size distribution, we believe this may provide an effective method of obtaining this data. Ongoing studies will determine if this device meets expectations. This program will be continued in 1994 if calibration studies are successful. A minimum of four sampling periods will be sampled and will consist of an early spring sampling period prior to smolt outmigration, an early summer period, a late summer period, and a pre-freeze up sampling period. These data will be coupled with sampling rearing juvenile sockeye salmon fry in the lakes by means of trawl net developed by Biosonics, Inc., capable of sampling differential depths. The collected fish will be sampled for AWL, lipid content, and stomach contents. Low abundances during the spring of 1993 resulted in collections of inadequate numbers of juvenile fish. Late fall collections along with early spring data will help to determine the relationship of temporal and spatial variation in abundances of sockeye fry and zooplankton prey with condition of sockeye fry and prior to the overwintering period.

Sampling will be performed along a subset of the transects established for fall fry fish sonar estimates in these lakes. Sampling depths will vary from the surface to the lower established limits of substantial zooplankton biomass. The species composition will be estimated using vertical net plankton tows representative of the area sampled with the optical plankton counter. Sample sizes will be determined by estimating the variance from a subset of the initial samples collected by the vertical tows. Species or life stage composition of zooplankton in the optical tow counts will be determined by comparing the length frequency data to those obtained from net samples or by apportionment.

In Skilak and Tustumena lakes at each of these four sampling periods, zooplankton and sockeye salmon diel migration will be estimated at one location. This will involve continuously sampling with the OPC over various depths and a concurrent hydroacoustic survey for one 24 hour cycle.

The above data will be integrated with the other information to develop a seasonal model of food availability within the euphotic zone of the lake to predict fish biomass production from these systems. The studies, begun in early spring, 1993, will continue through 1994 for an additional calendar year.

The holistic approach proposed here involves several evaluation procedures to assess the effects of sockeye salmon overescapement. First, fresh-water production from the 1989 escapements will be assessed in Kenai/Skilak, Red, and Akalura lakes. This will be accomplished through analysis of growth, freshwater survival (in particular over-winter survival), and freshwater age of sockeye smolt populations. Any anomalies will be determined by analysis of freshwater growth recorded on archived scales, historical freshwater age composition, and modelled freshwater survivals; and from results of previous studies as well as the smolt characteristics from each of the study systems. Also, planktonic food sources will be assessed through estimation of zooplankton prey biomass and diversity of species. Some of these analyses have been completed (Schmidt and Tarbox, 1993).

Losses of adult sockeye production from subsequent parent years may result from negative effects of progeny of the 1989 escapement on the lake's carrying capacity. The spawner/recruit relationships will be estimated from historical stock specific return data (where available), and generalized spawner/recruit data scaled to the carrying capacity parameters (i.e., euphotic volume and zooplankton biomass) of the nursery lakes where stock specific return data are not available (Geiger and Koenings 1991). If it is determined that in any of the affected systems, the density dependent effects are occurring outside of the traditional models, the effects will be isolated by examining a broader time window of the rearing life history of these species.

Third, experimental and empirical sockeye life history/production models (Koenings and Burkett 1987, Koenings et al 1989) will be used to compare salmon production by life-stage at escapement levels consistent with management goals to the 1989 escapements. These models will be refined by use of food availability

data obtained through the vertical sampling studies initiated in 1992.

Additionally, in the case of the Kenai system, the 1989 escapement effects will be viewed independently of the effects on previous brood years with high escapement.

Consult Schmidt and Tarbox (1993) for further discussion of analyses and methods used to date in progress reports on these investigations.

5. Location:

Study locations are on Kodiak Island and the Kenai Peninsula. Specific sampling locations are identified in Schmidt and Tarbox (1993).

6. Technical Support:

Administrative support is provided by the Administrative Division, Habitat Division, and Commercial Fisheries Management and Development Division staff of the Alaska Department of Fish and Game. The project leaders and their assistants are not funded by this project and are supported with general funds from the State of Alaska. Most laboratory analyses are conducted by the limnology laboratory in Soldotna. These studies are integrated with ongoing studies by the Commercial Fisheries Management and Development Division on Kodiak Island and the Kenai Peninsula. These studies have different objectives, i.e. to manage, enhance, and rehabilitate common property salmon fisheries, but use the same techniques and data collection methods. Consequently the EVOS investigations have been integrated into the normal operations of these Divisions for efficiency in completing the objectives of these studies and the general mission of these agencies.

NRDA STUDIES- OCTOBER 1993-SEPTEMBER 1994

	October	November	December	January	February	March	April	May	June	July	August	September
Kenai Stock												
Russell R. Stock												
Kasilof Stock												
Kasilof Hydro												
Skilak Hydro- pop est *a												
Skilak Hydro- diel dist												
Tustumena Hydro												
Skilak Growth												
Skilak Lipid												
Skilak Stomachs												
Kasilof Growth												
Kasilof Lipid												
Skilak Limno												
Kasilof Limno												
Skilak Diel Zoo												
Kasilof Diel Zoo												
Tustumena Diel Zoo												
Other Zoo												
Kenai Stock Acoustics												
Russell River Genetics												
Russell Lake pop est, twat, lim												
Kodiak Prince, Rod and Akahara												

Field activities

Construction, pre-season/post-season activities

Data analysis

Filename: 93-techd.v13
4/3/93

*aThe Skilak Hydro December work was proposed for 1994.

Figure 1. FFY94 Schedule

7. Contracts:

Technical support for specialized analyses are conducted by reciprocal service agreements with the University of Alaska at Palmer (lipid analysis) and Fairbanks (Nitrogen 15 analysis). These contracts were issued in 1992 and 1993 and will be issued in 1994-95, to provide specialized analysis not routinely used by the Limnology laboratory. These contracts were initiated after the project leader compared the cost effectiveness of internalizing the costs to the project and obtained price quotations for non-Alaska laboratories with the capability of conducting these analyses.

D. SCHEDULES

The Gantt Chart in Figure 1 outline the FY94 study plans. These plans are subject to minor modification following completion of data analysis during the winter of 1993-94.

E. EXISTING AGENCY PROGRAM

The Alaska Department of Fish and Game has ongoing commercial fisheries research operations on the Kenai and Kasilof River, Frazer Lake, Red River, Akalura Lake, Upper Station Lake, and Afognak Lake. In addition, the Division has ongoing data collection activities from Hidden, Karluk, and Spiridon lakes relating to the limnology of these systems. These data are integrated into statewide or regional data bases that are use to directly assess the impacts of the oil spill or are used as controls to measure the response of the studies proposed in this plan, against. In addition, the area research and management biologists for the Division of Commercial Fisheries management and development, the principal limnologist, the regional limnologist, and numerous administrative and support staff are supported by general funds provided by the Alaska legislature. To date, most of the data analysis and reporting for the Sockeye Salmon Over-escapement project has been provided for from contributions of the State of Alaska from these general funds. Total funding for these programs exceeds \$1 million. The following is a synopsis of projects in the areas that are covered by State of Alaska general funds:

Kodiak Island		
Kodiak Bio Rehabilitation	\$174.1	
Spiridon Lake Assessment-	\$42.8	
Frazer Lake /smolt operation	\$36.6	
Kodiak Finfish research	\$154.4	
Kodiak Frazer/Upper Station		
In-season forecasting	\$31.5	
Kodiak Major System Weirs-	\$168.2	
Total		\$607.6
Kenai Peninsula		
Statewide Limnology Staff (Part)	\$160	
Limnology & Estuarine Studies	\$100	

Cook Inlet Aquaculture Association		
Contracts (Hidden Lake-Others)	\$18	
Kenai River Sonar	\$45.4	
Kasilof River Sonar	\$28.7	
Soldotna Research Staff	\$203.1	
Stock Identification, Upper Cook Inlet	\$ 56.9	
Total		\$612.1
Grand Total		\$1219.7

F. ENVIRONMENTAL COMPLIANCE/PERMIT/COORDINATION STATUS

The studies proposed provide for data collection and field sampling programs. As such no environmental effect of these programs occurs beyond that of traditional fisheries management data collection activities and is within existing collecting permits or Federal special use permits issued to the Department of Fish and Game for scientific data collection activities. New programs on the National Wildlife Refuge are updated through permit amendments as needed. No other permits or other coordination activities are involved.

G. PERFORMANCE MONITORING

The performance of this project is monitored through the checks and balances of the State of Alaska accounting system within the Commercial Fisheries Management and Development, Habitat and Administration divisions of the Department of Fish and Game and the Department of Administration. Contractual compliance, personnel hiring, OEO compliance, and other administrative provisions are within the State of Alaska hiring and administrative chains of command and covered in standard operating procedures and administrative regulations. Replacement personnel are readily available by reassignment from over 30 permanent and seasonal staff within the Commercial Fisheries Management and Development Division of the Alaska Department of Fish and Game in Soldotna and Kodiak, when temporary problems are encountered. Filling new position follows state hiring guidelines when permanent vacancies occur. Project time frames for reports and analysis are maintained through proper planning and integration of these activities within the existing administrative structure of the Commercial Fisheries Management and Development Division. This project is one of over fifteen currently maintained by the limnology section and is administered accordingly. Quality control of the laboratory is conducted routinely following methods outlined in the laboratory manual referenced in this report. The laboratory is rated annually by the USGS nationwide laboratory rating system by conducting tests on blind samples provided by this group. Replicates are routinely run to cross check analytical techniques.

The scientific and technical aspects of the study are subject to internal review within Commercial Fisheries Management and Development Division. Publications

are submitted through an internal peer review process with the major findings submitted to peer review journals. Reports, work plans and study design are subject to the peer review process established by the EVOS Board of Trustees and Chief Scientist office. This year technical aspects of the studies findings to date and future plans were reviewed by a panel of international sockeye salmon researchers in a special half day session of the Kokanee and Sockeye Salmon workshop sponsored by the Northern Pacific International Chapter of the American Fisheries Society at Vancouver, B.C. in March, 1992. Interim annual status reports will be generated with publications being provided in peer review journals and scientific symposia as significant findings are obtained. The final report will be issued upon completion of the final year of field data collection.

H. COORDINATION OF INTEGRATED RESEARCH EFFORT

The investigations of Kodiak and Kenai River sockeye salmon have been integrated with long term research efforts by the Alaska Department of Fish and Game on these stocks. In addition, studies by the limnology laboratory and the fisheries development staff on Kodiak Island on these systems are included in data analysis. Study design and methodology builds off of earlier efforts. Planning and permitting of research activities and future rehabilitation efforts are coordinated through the USFWS Refuge staff in Soldotna and on Kodiak Island. Consultation and planning is conducted with the newly formed National Biological Survey Fisheries Research Laboratory staff in Anchorage. Development of restoration strategies on the Kenai Peninsula are through a review process with the regional planning teams and with an ADF&G review committee including the Sport Fish Division, when adjustments of management policies, such as escapement goals are involved. In addition, studies results from the Coghill Sockeye Salmon investigations in Prince William Sound are reviewed and integrated into the data analysis process for determining the response of the Kenai Peninsula ecosystem to restoration measures.

I. PUBLIC PROCESS

The investigations have been and will continue to be part of the public process. Reports have been prepared to the Alaska Board of Fisheries, the Upper Cook Inlet Regional Planning Team, various fishermen's organizations and civic organizations in Kodiak and on the Kenai Peninsula, in addition to the Trustee Council Public Advisory Group process. Scientific and technical review have included the peer review system established by the Trustee Council. In addition, technical aspects of the studies findings to date and future plans have been reviewed by a panel of international sockeye salmon researchers in a special half day session of the Kokanee and Sockeye Salmon workshop sponsored by the Northern Pacific International Chapter of the American Fisheries Society at Vancouver, B.C. in March, 1993.

J. PERSONNEL QUALIFICATIONS

Dana Charles Schmidt
Alaska Department of Fish and Game
Commercial Fisheries Management and Development Division
34828 Kalifornsky Beach Rd, Suite B
Soldotna, Alaska 99669
(907)262-9368

EMPLOYMENT:

October, 1991 to Present. Limnologist III, Principal Limnologist, FRED Division, Alaska Department of Fish and Game, Soldotna, AK. Responsibilities include establishing research objectives for the Statewide limnological investigations of the Commercial Fisheries Management and Development Division. This section provides direction for other components of the Division for determination of stocking rates for sockeye salmon in lakes and in the application of fertilization. This section also provides input to the commercial fisheries division for determination of the escapement goals for sockeye salmon. Supervise the limnology laboratory which completes water quality and plankton analysis for water samples taken from several hundred lakes statewide.

April, 1985 to October, 1991: Fishery Biologist IV, Regional Research Biologist, Westward Region, Alaska Department of Fish and Game. Responsible for establishing research objectives and priorities for the Westward Region Commercial Fisheries Division. This Division has management authority over extensive salmon and herring stocks on the Alaska Peninsula and Kodiak Island, in addition to management of the major shellfish stocks in the Gulf of Alaska and the Bering Sea. Annual ex-vessel value of these fisheries is several hundred million dollars. Research highlights included studies of crab larvae settling rates in the Gulf of Alaska and investigations on the effects of oil spill overescapement on the sockeye salmon production of major lakes on Kodiak Island.

May, 1982 to September, 1985 Acting F.B. IV, Susitna River Aquatic Studies Coordinator, Alaska Department of Fish and Game. The entire program under supervision included approximately 25 permanent and 50 seasonal employees. During this interim period, responsible for reorganizing the studies into a more efficient structure to meet the long term monitoring needs for determination of the effects of the Susitna project on the aquatic resources of the Susitna River. Supervised development of operational plans for 18 technical study programs on the Susitna River, assignment of priorities of tasks, and review of the technical merit of the programs proposed. Prior to January 1985. F.B. III, Resident and Juvenile Anadromous Project Leader, Su-Hydro Aquatic Studies Program, Alaska Department of Fish and Game. Supervised research programs on resident and juvenile anadromous fish in the Susitna River that may be impacted by

development of the Su-Hydro Project. Technical studies included development of models of sport fishery exploitation on arctic grayling populations, modelling instream flow responses of juvenile salmon habitat, development of baseline population parameters of resident fish and juvenile salmon and development of projections of supersaturated gas dissipation below the proposed dam sites.

January, 1981 to May, 1982: Fishery Biologist, Terrestrial Environmental Services, Anchorage, Alaska. Responsible for field and office review of the aquatic studies programs of the Alaska Power Authority for the Susitna Hydro-Electric Program. This responsibility included assisting the Alaska Department of Fish and Game in study plan development, providing preliminary assessment of impacts of the project on aquatic resources and presenting to the public progress of the aquatic studies programs.

May, 1980 to October 1980: Fishery Biologist, U.S. Fish and Wildlife Service, Soldotna, Alaska. Assisted on a radio-telemetry project and juvenile salmon habitat survey on the Kenai River, 6-mile Creek and the Deshka River in the Cook Inlet area. Activities included tagging and radio tagging chinook and coho salmon, collection of juvenile salmon and measurements of associated habitat, and assisting in the analysis of scale patterns from Kenai River chinook salmon. Other activities included statistical analysis of data, report review and preparation of a publication on the Kenai River chinook for Alaska magazine.

EDUCATION:

Ph.D. in Fisheries 1973

Major Field - Fisheries- Minor Field Pharmacology,

Oregon State University, Corvallis, Oregon

M.S. in Biology, 1970 Major Field - Aquatic Biology Minor Field - Sanitary Biology, University of Utah, Salt Lake City, Utah

B.S. in Wildlife Biology, 1968, University of Montana, Missoula, Montana

Ken Tarbox

Alaska Department of Fish and Game

Commercial Fisheries Management and Development Division

34828 Kalifornsky Beach Rd, Suite B

Soldotna, Alaska 99669

EMPLOYMENT:

May, 1980 to Present. Upper Cook Inlet Research Project Leader, Alaska Department of Fish and Game, Soldotna, Alaska. Responsibilities include planning, implementing, supervision, and reporting on various salmon related research and management projects. These involve hydroacoustic enumeration of salmon in glacial systems, defining salmon migratory behavior in both salt and fresh water, evaluation of potential impacts of

resource development on habitat and populations, management of the UCI commercial salmon fisheries, stock identification studies using scale or genetic markers, and life history studies of sockeye salmon.

March, 1972 to May, 1980. Project manager and Senior Biologist, Woodward Clyde Consultants, Anchorage, Alaska. Responsibilities included supervision and research for a number of projects. These included an evaluation of existing methodologies for determining instream flow requirements for Alaskan fishes, determining the biological impact of a dredging projects located in lower New York Harbor and Lake Michigan, fishery investigations in the Zayandeh River, Iran, impact assessment of various oil related projects in Virginia, North Carolina, Texas, and Prudhoe Bay, Alaska, and studies and evaluation of impacts associated with nuclear power plants in New Jersey, Louisiana, Indiana, and Pennsylvania.

July, 1970 to March, 1972. Research Assistant, Louisiana Co-operative Fishery Unit, Louisiana State University, Baton Rouge, La. Responsibilities included the design and conduct of a one year investigation of juvenile fish behavior in an estuarine environment.

EDUCATION:

M.S. in Fisheries, 1974. Louisiana State University, Baton Rouge, La.
B.S. in Fisheries Science. 1970. University of Washington, Seattle, Wa.

CERTIFICATIONS:

Fisheries Scientist, Certificate 1165, American Fisheries Society, 1976.

Stan R. Carlson
Alaska Department of Fish and Game
Commercial Fisheries Management and Development Division
34828 Kalifornsky Beach Rd, Suite B
Soldotna, Alaska 99669

EMPLOYMENT:

January 1993 - present: Biometrician for the Alaska Department of Fish and Game, Limnology Section, Commercial Fisheries Management and Development Division, Soldotna, Alaska. Supervised by Dr. Dana Schmidt. Conduct statistical data analyses to evaluate factors that affect dynamics of the biota in lake ecosystems. Design limnological experiments and determine methods to estimate zooplankton and salmon abundance. Develop and approve methods to estimate hatchery contributions to the fishery. Develop, review, and conduct statistical analyses for projects related to the impact of oil on commercial fishery species. Provide biometrical consulting to area and regional biologists and statewide limnologists.

November 1991 - January 1993: Mathematical Statistician for the National Marine Fisheries Service, Auke Bay Laboratory, Juneau, Alaska. Supervised by Mr. Steven Ignell. Conduct statistical studies on community attributes of pelagic fauna in the north Pacific Ocean. Provide biometrical consulting, technical editing, and collaborative input on projects such as salmon bycatch and climate change studies.

January 1989 - May 1991: Statistics Teacher, Experimental Statistics Department, New Mexico State University, Las Cruces. Supervised by Dr. Michael Ames. Instruct laboratory courses in statistics for undergraduate science majors.

May - August 1990: Research Specialist (statistician), Department of Entomology, Plant Pathology, and Weed Science, New Mexico State University. Dr. Ellis Huddleston, Supervisor. Provide statistical modeling, analysis, and design of experiments related to agricultural field studies and pest management programs.

May - December 1988: Field Biologist, Biology Department, New Mexico State University. Supervised by Mr. Roger Skaggs. Conduct field population surveys and habitat analyses of night birds in Lincoln National Forest, New Mexico. Collect field data, supervise field personnel, and maintain data records. Develop operational strategies and conduct follow-up statistical estimation procedures.

August 1985 - June 1988: Graduate Assistant, Biology Department, New Mexico State University. Supervised by Dr. Ralph Raitt and Dr. Walt Whitford. Teach undergraduate biology and zoology laboratory courses. Collect data and maintain field ecology experiments for ecological research programs. Develop and conduct original field research on desert insect ecology.

June 1983 - May 1985: Research Specialist, Gordon Environmental Studies Laboratory, University of Montana, Missoula. Supervised by Dr. Philip Tourangeau. Manage data, conduct quality assurance/control procedures, and perform statistical analyses for environmental science projects. Aid in the design and implementation of field research, primarily in the area of pollution biomonitoring.

EDUCATION:

1991 Master of Science, Experimental Statistics, New Mexico State University.

1988 Master of Science, Biology (ecology), New Mexico State University.

1983 Bachelor of Arts, Environmental Biology, University of Montana.

K . BUDGET

ADF&G

Personnel	602.8
Travel	9.7
Contractual	71.2
Commodities	53.3
Equipment	22.5
Capital Outlay	<u>0.0</u>

Sub-total	759.5
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General Administration	95.4
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Project Total	854.9
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NEPA Compliance	0(State contributed)
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Literature Cited:

Enright, J.T. 1977. Diurnal vertical migration: Adaptive significance and timing. Part 1. Selective advantage: a metabolic model. *Limnol. Oceanogr.* 22, 856-872.

Geiger, H. J., and J. P. Koenings. 1991. Escapement goals for sockeye salmon with informative prior probabilities based on habitat considerations. *Journal of Fisheries Research* (in press).

King, B.E., L. K. Brannian, and K. E. Tarbox. 1991. Kenai River sockeye salmon smolt studies, 1990-91. Alaska Department of Fish and Game Division of Commercial Fisheries Regional Information Report No. 2S91-8, Anchorage.

Koenings, J. P. and G. B. Kyle. 1991. Collapsed populations and delayed recovery of zooplankton in response to heavy juvenile sockeye salmon (*Oncorhynchus nerka*) foraging. (Proceedings: International Symposium on Biological Interactions of Enhanced and Wild Salmonids held at Nanaimo, B. C., Canada). *Spec. Publ. Can J. Fish. and Aquat. Sci.*

Koenings, J. P., and R. D. Burkett. 1987. Population characteristics of sockeye salmon (*Oncorhynchus nerka*) smolts relative to temperature regimes, euphotic volume, fry density, and forage base within Alaskan Lakes. p. 216-234. *In* H. D. Smith, L. Margolis, and C. C. Wood [ed.] Sockeye salmon (*Oncorhynchus nerka*) population biology and future management. *Can. Spec. Publ. Fish. Aquat. Sci.* 96.

Koenings, J. P., J. E. Edmundson, G. B. Kyle, and J. M. Edmundson. 1987. Limnology field and laboratory manual: Methods for assessing aquatic production. Alaska Department of Fish and Game, FRED Division Report Series No. 71:212 p.

Koenings, J. P., R. D. Burkett, M. Haddix, G. B. Kyle, and D. L. Barto. 1989. Experimental manipulation of lakes for sockeye salmon (*Oncorhynchus nerka*) rehabilitation and enhancement. Alaska Department of Fish and Game, FRED Division Report Series No. 96:18p.

Kyle, G. B., J. P. Koenings, and B. M. Barrett 1988. Density-dependent, trophic level responses to an introduced run of sockeye salmon (*Oncorhynchus nerka*) at Frazer Lake, Kodiak Island, Alaska. *Canadian Journal of Fisheries and Aquatic Sciences* 45:856-867.

Kyle, G. B. 1992. Summary of sockeye salmon (*Oncorhynchus nerka*) investigations in Tustumena Lake, 1981-1991. Alaska Department of Fish and Game, Division of Fisheries Rehabilitation, Enhancement and Development Division Report No. 122, Juneau.

Kyle, G. B. 1989. Summary of acoustically-derived population estimates and distributions of juvenile sockeye salmon (*Oncorhynchus nerka*) in 17 nursery lakes of Southcentral Alaska. Alaska Department of Fish and Game, FRED Division Report Series No. (In review).

Kyle, G. B. 1983. Crescent Lake sockeye salmon smolt enumeration and sampling, 1982. Alaska Department of Fish and Game, FRED Division Report Series No. 17:24 p.

Mills, E. L., and A. Schiavone, Jr. 1982. Evaluation of fish communities through trophic assessment of zooplankton populations and measures of lake productivity. North American Journal of Fisheries Management 2:14-27.

Pearre, Sifford Jr. 1979. Problems of detection and interpretation of vertical migration. J. Plank. Res. 1(1):29-44.

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Schmidt, D. C. and K.E. Tarbox, 1993. Sockeye salmon overescapement. State/Federal Natural Resource Damage Assessment Status Report. Fish/Shellfish Study No. 27. FRED Tech. Rept No. 126.

Tarbox, K.E., and B.E. King. 1989. An estimate of juvenile fish densities in Skilak and Kenai Lakes, Alaska through the use of dual beam hydroacoustic techniques in 1989. Alaska Department of Fish and Game, Commercial Fish Division Regional Information Report No. 2S90-1.

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EXXON VALDEZ TRUSTEE COUNCIL
1994 Federal Fiscal Year Project Budget
October 1, 1993 - September 30, 1994

Project Description: Sockeye Salmon Overescapement - This project is a continuation of 93002 and monitors sockeye salmon smolt outmigration and the density of fry rearing in the nursery lakes of the Kenai Peninsula, Kodiak Island, and the Chignik system on the Alaska Peninsula. In addition, detailed investigations will be conducted to determine both abiotic and biotic factors that may have contributed to the decline in production.

Budget Category:	1993 Project No. 93002 Authorized FFY 93	'93 Report/ '94 Interim* FFY 94	Remaining Cost** FFY 94	Total FFY 94	FFY 95	Comment
Personnel	\$584.0	\$290.1	\$312.7	\$602.8	\$375.0	Report: \$180.0
Travel	\$11.5	\$13.5	\$0.0	\$13.5	\$10.0	\$10.8
Contractual	\$113.3	\$18.3	\$52.9	\$71.2	\$75.0	\$11.4
Commodities	\$70.8	\$12.3	\$37.2	\$49.5	\$40.0	\$8.0
Equipment	\$56.6	\$0.0	\$22.5	\$22.5	\$30.0	\$0.0
Capital Outlay	\$0.0	\$0.0	\$0.0	\$0.0	\$0.0	\$0.0
Subtotal	\$836.2	\$334.2	\$425.3	\$759.5	\$530.0	\$210.2
General Administration	\$93.9	\$44.8	\$50.6	\$95.4	\$61.5	\$27.8
Project Total	\$930.1	\$379.0	\$475.9	\$854.9	\$591.5	\$238.0
Full-time Equivalents (FTE)	11.8	5.3	5.7	11.0	6.7	FFY 95 costs may vary depending upon the outcome of the FFY 94 results.
Dollar amounts are shown in thousands of dollars.						
Budget Year Proposed Personnel:		Reprt/Intrm	Reprt/Intrm	Remaining	Remaining	
Position Description		Months	Cost	Months	Cost	
Reprt	7 Kodiak Commfish Positions	9.0	\$39.4	8.5	\$43.7	
	2 Kodiak Development Positions	6.0	\$26.4	1.5	\$7.4	
	9 Kenai Limnology Positions	24.0	\$119.5	18.0	\$92.3	
	24 Kenai Commfish Positions	24.0	\$101.4	32.0	\$134.8	
	5 Tustemena Limnology Positions	0.0	\$0.0	7.0	\$27.8	
Intrm	1 Program Manager	0.5	\$3.4	1.0	\$6.7	
Personnel Total		63.5	\$290.1	68.0	\$312.7	NEPA Cost: \$0.0
						*Oct 1, 1993 - Jan 31, 1994
						**Feb 1, 1994 - Sep 30, 1994

07/14/93

1994

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Project Number: 94258
Project Title: Sockeye Salmon Overescapement
Agency: AK Dept. of Fish & Game

FORM 2A
PROJECT
FILE

EXXON VALDEZ TRUSTEE COUNCIL
 1994 Federal Fiscal Year Project Budget
 October 1, 1993 - September 30, 1994

Travel:		Reprt/Intrm	Remaining
Reprt	12 RT Soldotna/Anchorage @ \$150 + 1 day per diem	\$3.6	\$0.0
	8 RT Soldotna/Juneau @ \$450 + 1 day per diem	\$4.9	\$0.0
	6 RT Soldotna/Kodiak @ \$300 + 1 day per diem	\$2.3	\$0.0
Intrm	4 RT Soldotna/Anchorage @ \$200/trip + 1 day per diem @ \$150/day	\$1.4	\$0.0
	2 RT Soldotna/Kodiak @ \$500/trip + 1 day per diem @ \$150/day	\$1.3	\$0.0
Travel Total		\$13.5	\$0.0
Contractual:			
Reprt	Water quality analyses	\$4.0	\$0.0
	Species Identification of plankton and macroinvertebrate samples	\$7.4	\$0.0
Intrm	Contract for air charters for 57 hours @ \$350/hour	\$6.9	\$11.6
	Freight	\$0.0	\$2.0
	Outboard motor and radio repair	\$0.0	\$3.0
	Chemical analysis of water samples collected in winter sampling	\$0.0	\$15.8
	External lipid and N15 contracts for fish tissue analysis	\$0.0	\$20.0
	(Lipids \$20/sample - 500 samples; N15 \$100/sample - 100 samples)		
	Software licensing	\$0.0	\$0.5
Contractual Total		\$18.3	\$52.9

07/14/93

1994

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Project Number: 94258
 Project Title: Sockeye Salmon Overescapement
 Agency: AK Dept. of Fish & Game

FORM 2B
 PROJECT
 DETAIL

EXXON VALDEZ TRUSTEE COUNCIL
 1994 Federal Fiscal Year Project Budget
 October 1, 1993 - September 30, 1994

Commodities:		Reprt/Intrm	Remaining
Reprt	Office and photo supplies, publication and binding cost	\$8.0	\$0.0
Intrm	Field supplies	\$4.3	\$0.0
	Scientific collection equipment (Limnology water samplers, bottles, nets, preservative, etc.)	\$0.0	\$19.3
	Outboard fuel and oil	\$0.0	\$2.0
	Boat/motor repair and maintenance	\$0.0	\$10.2
	Groceries (10 people x 20 days x \$20/day)	\$0.0	\$4.0
	Safety supplies (fire extinguishers, float coats, life jackets)	\$0.0	\$1.0
	Foul weather gear	\$0.0	\$0.7
Commodities Total		\$12.3	\$37.2
Equipment:			
	Boat and motor (1 boat - 2 motors)	\$0.0	\$10.0
	2 outboard motors	\$0.0	\$8.0
	Fry tow nets, smolt trap	\$0.0	\$2.0
	Computer for field data entry aboard vessel	\$0.0	\$2.5
Equipment Total		\$0.0	\$22.5

07/14/93

1994

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Project Number: 94258
 Project Title: Sockeye Salmon Overescapement
 Agency: AK Dept. of Fish & Game

FORM 2B
 PROJECT
 FILE