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Exxon Valdez Oil Spill
Coastal Habitat Project
Herring Bay Experimental Field Station
1991 Field Experiments:
Restoration Monitoring and Feasibility

STUDY ID NUMBER:

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LEAD AGENCY: US Forest Service

COST OF PROPOSAL: \$245,000.00

INCLUSIVE DATES OF STUDY PLAN: March 1, 1991-February 29, 1992

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II. INTRODUCTION:

As a part of the Coastal Habitat program, the US Forest Service, through the University of Alaska Fairbanks (UAF), established an experimental field station in Herring Bay, Knight Island, during 1990. The purpose of the station is to provide a research platform for intertidal Natural Resource Damage Assessment (NRDA) and Restoration-related studies.

During the summer of 1990, UAF implemented separate studies on 15 pairs of oiled and non-oiled sites in Herring Bay. These ongoing studies have initially applied to assessment of damages; however, four studies are directly relate to the feasibility of intertidal restoration and monitoring of natural recovery.

One set of studies examines presence/absence differences in population dynamics between common intertidal species on impacted and reference sites. Results from these studies will summarize a checklists of species, and compare differences and densities per unit area.

A second group of studies examines settlement between oiled and non-oiled surfaces. Results from these studies compare density information of each species examined, per unit area, between oiled and control sites. A third study examines succession of algae at 104 study plots in Herring Bay.

A forth group of studies consists of a series of experiments which will monitor the ability of the dominant intertidal macrophyte, *Fucus*, to recover from the effects of the oil spill and/or spill clean-up efforts in Herring Bay. In addition we will be monitoring all algal species in the intertidal zone for rates of natural recover from spill damage.

For NRDA, these studies will provide immediately useful data regarding impacts associated with the Exxon Valdez spill. The continuance of these and other monitoring programs in 1991 will contribute to the base of knowledge concerning restoration feasibility, especially with respect to determining a rate of recovery to pre-impacted conditions.

III. OBJECTIVES:

1. Compare between oiled and control sites abundance per unit area of intertidal invertebrates with limited dispersal capabilities. Based upon the data generated in 1990, compare 1991 field data between oiled and control site pairs. If differences between replicated site pairs are showing signs of a "normalization" (or no significant differences between populations), then forecast a "rate" at which populations might be expected to be similar.

2. Compare the success of settlement by barnacles and algae (e.g. Fucus sp.) between oiled and non-oiled substrates. Based upon data generated from the 1990 and 1991 seasons, estimate the length of time required before oiled substrates show no significant differences in colonization success by barnacles.
3. At the termination of the limpet grazing experiment in the 1990 field season, an opportunity was presented to gain an understanding of temporal algal succession within Herring Bay. Several questions regarding algal succession in general are relevant to possible impacts from oil. These questions include:
 - A. Can a basic algal succession for Herring Bay be defined?
 - B. Based upon the number of sites chosen for study, is there a difference in algal succession (and thus, species composition) between reference and impacted sites?
 - C. Is there a difference in algal succession between areas that are simply scraped free of all algae, vs. the application of a killing agent (such as bleach)? *also correct*
4. Monitor the natural restoration and recovery of the intertidal algae, with special emphasis on Fucus in Prince William Sound. Sub-objectives are:
 - A. The relative fecundity of Fucus plants will be determined to ensure that the existing populations have the capacity to "re-seed" the damaged areas.
 - B. Fucus population dynamics will be monitored in oiled and unoiled areas in order to assess natural restoration rates.
 - C. Algal recolonization and Fucus growth rates will be measured to assess how long it may take for the intertidal algal species to recover to pre-spill levels.

IV. METHODS:

This section defines each study conducted at the experimental station in 1991, and identifies the methods used. Standard Operating Procedures for these studies are listed as Appendix 1 & 2 under OTHER INFORMATION.

A. Sampling Methods:

Site Characterizations

Study sites are characterized through the establishment of three random transects along the site perpendicular to the water line, starting at Mean High High Water (MHHW). Quadrats were located randomly within the first three meters of vertical fall along each transect. In each quadrat, presence/absence data for all invertebrates and algae were recorded, as well as determination of percent cover. Also, data for temperature and salinity were collected at each of the study sites on a weekly basis. Refer to the 1991 SOP in item X for modifications to this study.

1. Population Dynamics of certain invertebrate species

This study has been designed to examine the differences in numbers and recruitment of certain invertebrates with limited dispersal capability, between oiled and non-oiled sites. Limpets are included in this monitoring study because of their likely importance as grazers to community structure.

Materials and Methods, Population Dynamics:

Permanent plots were established at five pairs of sites: three sheltered rocky and two pairs of sheltered coarse grained environments. These plots were established at three meters of vertical fall along six randomly placed transects across the site length, establishing a total of 18 study plots per site. Quadrat dimensions were 20 X 50 cm. Within each of these permanent plots, all limpets, Nucella spp., Littorina sitkana and Leptasterias hexactis were counted. Also, using a 1 m semicircle adjacent to and centered at the left of the 20 X 50 cm quadrat, the nearest of each of these species was measured and recorded. Refer to the 1991 SOP in item X for modifications to this study.

2. Settlement Studies

A. Barnacles

Within Herring Bay, certain oiled shorelines still possess heavy accumulations of dried tar, especially in the upper intertidal zone, where desiccation and baking by sunlight has resulted in an asphalt condition of the oil. Established colonies of barnacles were obviously impacted along many of these areas. A study was implemented which examined whether the presence of such tar reduces the settlement capability of cyprid barnacles relative to cleaned areas within a tarred substrate.

Materials and Methods, Barnacles:

Two oiled sites and two reference sites of similar character were selected in Herring Bay for this study. Sites 1641A and 1342D are oiled vertical faces located on the southern end of a small island, in the lower center of Herring Bay. All sites have vertical faces where barnacles presently exist, or in the case of sites that were heavily oiled and treated, having many skeletons still attached to the substrate. Sites 1641B & 1642C are non-oiled reference sites in the southeastern cove of Herring Bay. All sites had high densities of the barnacle Semibalanus balanoides.

At each site, paired 10 X 10 cm plots were established. One member of each pair was scraped and brushed to remove all visible tar (or barnacles in the cases of the non-oiled sites). The length of each site was measured, and the number of planned pairs divided into the site length. The first plot was placed randomly, within the first segment, and subsequent plots were placed at equal distances from the first. A coin was flipped to determine which 100 cm² area of the first pair to scrape. The subsequent scraped plots were then alternated.

The sites were periodically examined for barnacle settlement, as well as germlings of the alga Fucus gardneri. The number of barnacle juveniles and germlings were recorded during each inspection. Each 100 cm² area was also photographed. Refer to the 1991 SOP in item X for modifications to this study.

B. Settlement on oiled and non-oiled substrates

A second study also examined differences in settlement of marine invertebrates and algae between oiled and non-oiled substrates. However, the substrates used in this second study were rocks retrieved from an oiled shoreline in Herring Bay, as well as rocks treated with fresh North Slope crude oil, taken from the T/V Exxon Valdez last year. The objectives of this experiment were to examine differences in: a) the percent cover of barnacles and macro algae; b) the number of individuals per unit area; and c) the presence/absence of invertebrate species on oiled and non-oiled substrates placed within various sites.

Materials and Methods, Substrate Transplants:

On a beach lying along the western arm of Herring Bay, seventy two oiled rocks of similar size were collected and returned to the laboratory. These rocks represent a substrate coated with 1 year old Exxon Valdez Prudhoe Bay Crude (EV). All rocks were collected and packed in boxes and separated by aluminum foil, so

that the rocks would not touch one another.

Upon return to the laboratory, all rocks were laid out and one-half of each rock was cleaned with the solvent, Methylene Chloride (MeCl_2), with the exception of 8, which were left completely oiled to serve as strata for a weathering analyses of the oil.

After each half was thoroughly cleaned, the rocks were allowed to dry. "Top" and "bottom" of each rock was determined with regard to symmetry and morphology. The "top" was assigned to the surface with the least irregularity. When dry, each rock received a unique identification number and was marked with an indelible marker. Each rock was measured with calipers for total length, and length of the cleaned and oiled sides, and each rock was then photographed.

Also, 72 rocks of approximately the same size were collected from a geologically similar, but un-oiled beach. Half of each rock surface was dipped in fresh Prudhoe Bay Crude (PB) until a "tarred" coating was achieved. These rocks were allowed to dry and were handled in a manner identical to the EV rocks.

In addition to the rocks, 72 clay tiles were incorporated into the experiment. The tiles, being uniform in surface texture and aspect, served as substrate heterogeneity controls for the rocks. Thirty six of these clay tiles were oiled with fresh PB oil and the other 36 remained clean. The tiles were placed side-by-side in the field as oiled and un-oiled pairs.

At each of the experimental sites, rocks and tiles were placed randomly at the 2 m elevation contour. Control rocks (i.e. rocks which were un-oiled, but had half of the surface treated with MeCl_2) were also placed at each site to test for use of the MeCl_2 solvent. Each site received an identical number of rocks and tiles representing the following experimental conditions:

The basic experimental unit has been left in the field indefinitely, and consists of 3 EV rocks, 3 PB rocks and six pairs of tiles. The additional rocks were placed to be destructively sampled at three separate time periods. These time periods were mid summer 1990, early fall 1990, and mid spring, 1991.

After placement of all substrates in the field, settlement by barnacles and macro algae on each surface was recorded. Counting involved use of a 3 cm X 3 cm quadrat. The quadrat edge was placed at the midpoint of the line separating the oiled and un-oiled portions of the rock. Where possible, individual species were identified, counted and recorded. Rocks were photographed at a fixed focal length to incorporate the quadrat.

Throughout this substrate transplant experiment, the chemical composition of crude oil will change over time. Consequently, the "thickness" of oil coating of the substrates will gradually decrease. Thus, a procedure was developed to quantify a rate of change in the oil's character. The procedure employed a gravimetric analysis of an area of oil extracted by MeCl_2 . Therefore, completely oiled EV and PB rocks were also placed in the field as controls for taking samples for this oil "weathering" analysis.

The oil weathering analysis entailed a MeCl_2 extraction of a 3 X 3 cm area on each of the control rocks, using a pre-weighed absorbent material. This absorbent material was then placed in a pre-weighed vial. Each vial was opened and stored at room temperature, and allowed to dry. The absorbent material was then reweighed. The sample vials were refilled with MeCl_2 , and refrigerated for Gas Chromatography/Flame Ionization Detection analysis (GC/FID). Refer to the 1991 SOP in item X for modifications to this study.

3. Algal Succession:

Because eight study sites were chosen for studies of limpets grazing (four pairs of impacted and reference sites) and the termination of the experiment involved removal of the fences and all algae within each 625 cm area, a platform was provided to easily examine algal species succession at multiple plots.

At the termination of the limpet grazing study, fences from two elevation contours were removed. Only the marine epoxy used to hold the fences in place was left to serve as a marker for each of the algal succession study plots.

All algae was removed from each 625 cm area. A coin was flipped to determine if the odd or even numbered fences would receive application of sodium hypochlorite (bleach).

Sessile invertebrates, such as barnacles were not removed from the 625 cm area. A photograph of each experimental was taken (with photolabels in each frame).

Each study plot will be revisited to assess algal species composition. At the time of visitation, percent cover of each plot by separate species will be determined by a point grid method. Also, photographs of each plot will also be taken, and a voucher specimen of each plant observed within each plot will be collected. Voucher specimens will be sent to UAF for taxonomic identification. Finally, numbers and species of invertebrate

grazers found within the 625 cm area will also be counted.

This process will be repeated until four time series plots have been established for both the scraped and bleached plots at each contour. These study plots will be monitored quarterly.

4. Fucus and algae Recovery

Experiment 1 - Fucus Reproductive Potential and Egg Viability

The Exxon Valdez oil spill may have affected the ability of the intertidal plants to produce sufficient, viable reproductive cells to repopulate the natural habitat. This experiment will assess the relative fertility of Fucus in oiled and control sites by measuring the rate of egg release from randomly selected receptacles. In addition the viability of the released eggs will be monitored. Because of the many factors that can affect the release of eggs from the conceptacles, this experiment must be conducted at the field camp to help reduce the variation in the data. This is a continuation of experiments conducted this past field season.

Plants for this study will be collected from the top three meters at each site. The same sites and plots will be used for this experiment as those used for the population dynamics study. Two semicircles of 0.5 m radii will be used to select two plants at each 20 cm x 50 cm quadrat. There will be a maximum of 36 plants selected at each site (2 per quadrat x 6 quadrats per tidal range x 3 tidal ranges). The left semicircle will have its origin (point from which the radius is extended) on the left edge of the 20 cm x 50 cm quadrat, 25 cm down from the top edge. The semicircle will be concave to the quadrat. The right semicircle will be a mirror image of the left semicircle, with its origin on the right edge of the quadrat, 25 cm below the quadrat marker on the transect line. To select each plant, a search will be made in each semicircle for the Fucus plant which is nearest to the origin of the semicircle and which has receptacles. Receptacles are defined as inflated tips of the Fucus branches which contain conceptacles (note that the conceptacles may be in varying stages of development). If no plants within the semicircle contain receptacles, then a zero will be recorded for the number of eggs released for the sample. At each subsequent sampling, two new semi-circles will be used which will be located along the same contour on each side of the quadrat and displaced 0.5 m farther from the origin of the last sampled semicircles in directions away from the quadrat.

The selected plants are removed intact from the substrate and placed in numbered plastic bags. Plants should be kept separated by site location and tidal range. Plants should be collected from the oiled and its paired control sites on the same day.

At the laboratory one receptacle is selected from each plant by

the following method. An arbitrary receptacle is selected on the plant, and a random integer between 1 and 10, inclusive, is generated on a calculator. Using this receptacle as number '0', count out the number of adjacent receptacles corresponding to the selected random integer. When counting, proceed in one direction, counting neighboring receptacles on neighboring branches. Count only receptacles and not sterile branch tips. The receptacle with the number corresponding to the random integer is the selected receptacle for egg release. Trim away the non-selected part of the plant, leaving only the receptacle. Blot this receptacle dry with paper towels then rinse for a few seconds with fresh water. Re-dry the receptacle thoroughly and place between dry paper towels. Set at 8-10 C in the dark for 24 hours. After weighing the receptacle to 0.01 g, immerse it in 15 ml of cold, sterile seawater in a 60 x 15 mm petri dish. Cover the dish and set it in an incubator at 8-10 C, with a photoperiod of 16:8 (L:D), at 50-80 micromoles/m²/s light.

After 48 hours remove the receptacle and return the dish to the incubator. After an additional 48 hours, remove the dish from the incubator and add Calcofluor solution, followed by 5% formalin 30 minutes later. Prepare the dish for shipment to JCFOs where the eggs will be counted and analyzed according to the procedure published in the CHIA SOP.

This experiment will be performed at three paired oiled and control rocky protected and two paired coarse textured sites in Herring Bay.

Experiment 2 - Fucus Population Dynamics

The population structure of the intertidal is often affected by disturbances. A disturbance such as a former clearing is evidenced by a preponderance of smaller plants in the population. This experiment will monitor the population structure of Fucus as it is affected by oiling and/or clean-up efforts. The plots selected for this experiment will be the same as those used for the recruitment studies in the intertidal invertebrate studies at Herring Bay. The sites include 4 sheltered rocky oiled sites and their matched control sites and 2 coarse gravel oiled sites with their paired controls. There are 6 transects per site with 3 quadrat locations per transect. The procedure for the location of these transects and quadrats is identical to that used in the stratified sampling done in the Comprehensive Assessment of Injury to Coastal Habitats surveys. However, only one 20 cm x 50 cm quadrat will be used at each quadrat location and will be placed with the upper right-hand edge of the quadrat located at the quadrat location point on the transect line. This experiment is a continuation from last field season.

At each quadrat, size-frequency distribution will be determined by measuring all visible Fucus plants for total length without

removing the plants from the substrate. For each plant the reproductive status and the condition of the plant will be recorded using the scheme outlined for the intertidal plant transect studies. Percent cover of all organisms, bare rock, and oil will be estimated with a 50-point grid placed over the quadrat. Drift algae will be removed before any data are taken. If necessary, a small sample of the surface oil will be removed with a knife and examined under a microscope to ensure that it is actually oil. The measurements will be repeated at least 3 times during the summer field season. All plots will be photographed at all sampling dates.

Experiment 3 - Fucus Germling Growth

This experiment, slightly modified from that performed last year will be conducted in four segments established along a transect line extending along the contour of the site at the 1.0 and 2.0 m of vertical fall, at the top of the Fucus zone. The experiment will measure the growth rates of Fucus germlings in oiled and unoiled sites. Small settling plates will be seeded at JCFS with Fucus eggs at an average density of 144 eggs per square cm. These plates will be shipped to Herring Bay and paired seeded and unseeded control plates will be placed in random order at the first and second meter vertical drop at three pairs of oiled and control sites. Separate sets of plates will be deployed at 4 to 6 week intervals.

Fucus plants, >1mm, growing on the plates will be photographed, enumerated and measured in the field to assess recruitment and growth throughout the summer. At the end of the summer, the plates will be photographed, removed from the rock surfaces, and plants will be enumerated and measured in the lab.

Detailed SOP's will be necessary for all aspects of this field work and will be generated during the month of March prior to the field season. The SOP's will be similar to those developed last year and published as part of the operational plan for the Coastal Habitat Injury Assessment project.

Temperature and Salinity:

Stations adjacent to all 1990 study sites were monitored weekly for temperature and salinity at the 0, 0.5 and 1 through 5 m depth contours around MHHW, using a CTD meter. This procedure will be continued through the 1991 season.

B. Citations:

Dayton, P. K. 1971. Competition, disturbance and community organization: the provision and subsequent utilization of space in a rocky intertidal community. Ecol. Monogr. 41:351-389.

While it is probable that many components of these methods have been employed by other researchers previously, no additional citations were specifically referred to in developing the above procedures.

C. Standard Operating Procedure Requirements:

As identified at the beginning of this section, the SOP's for the experimental field station are presented as appendices 1 & 2 under item X, OTHER INFORMATION.

D. Quality Assurance and Control Plans.

1. Data Collection and Analysis.

For each of the studies identified in this plan, detailed data collection forms have been developed (see Appendix 1 and Section VI-B, Sample and Data Archival). The original, completed forms are maintained in a notebook while at the experimental station. These same notebooks are then stored at the University upon termination of each field season. In addition to the field data forms, the experimental station maintains the field data in an automated database. The contents of this database are also transferred to the UAF data management department.

2. Sample Collections and Labelling.

All hydrocarbon samples collected by the experimental field station in Herring Bay, are done so according to the sample collection procedures and chain-of-custody requirements discussed in the Analytical chemistry group technical services document #1, NMFS, Auke Bay, Alaska, entitled "State/Federal damage assessment plan analytical chemistry collection and handling of samples". Other samples (i.e. invertebrates) not used for hydrocarbon analyses are collected at the experimental station for voucher purposes. These samples are maintained at the experimental station, and transported to UAF upon termination of the field season.

V. DATA ANALYSIS:

The experiments described in this plan have been designed largely for standard parametric and non-parametric statistical analyses (i.e. Student's T-test, Wilcoxin Rank sum test, Sign test, Mann Whitney U-test, analysis of variance). Replication in the selection of study site pairs is the basis for analytical power.

The products to be generated from these experiments will be in digital and non-digital form. Report results will include graphics in the form of maps, graphs, tables and figures.

VI. SCHEDULES AND PLANNING:

A. Data and Report Submission Schedule.

Data collection and information management will be conducted simultaneously while at the field station. Because the station is computer-equipped, data entry will be performed routinely. Consequently, the availability of raw data will be immediate upon termination of the 1991 field season. Report writing and submission will be completed within 60 days of the sampling season's termination.

B. Sample and Data Archival.

The sample and archival system for this research is documented as follows:

1. This study Plan, with all approved revisions
2. All SOPs related to the Herring Bay experimental field station are included in appendices 1 & 2
3. A complete set of 1990 field data from the Herring Bay Experimental field station is on file at the University of Alaska Fairbanks. Sediment hydrocarbon samples, and limpet tissue hydrocarbon samples are stored at the Auke Bay Marine Laboratory as part of the hydrocarbon sample pool
- 4&5. All records, logs, summaries, and reports used during 1990 are also on file at the University of Alaska Fairbanks. Samples of the data log sheets used for each study are provided as Appendix 3, under item X.

C. Management Plan

The management plan for the experimental field station is straight forward. The sampling season will run from April 15 to September 15, 1991. Refer to the Logistics section below for a discussion of facilities. As most experiments described here are in place, their frequent monitoring is the primary requirement. Five resident field staff will monitor all invertebrate studies. Workable tides for these studies lie around the 0.0 tide level from MHHW. There are brief periods during each month where adequate low tides and daylight will not correspond. Each study site will be monitored twice weekly, on average, unless otherwise specified in the SOP.

D. Logistics

The presence of the experimental field station within Herring Bay

makes access to the study sites ideal. This station (a floating barge) is currently in place and permanently moored within safe harbor in Herring Bay, and meets all Coast Guard safety regulations. Most of the field equipment is already on board. Initiating the 1991 sampling season will require transport of personnel and some equipment to the station. Throughout the season, the facility will receive food and supplies on a weekly basis. All study sites are located within Herring Bay, and are accessed via small gas-powered skiffs.

VII. BUDGET: (in thousands of dollars)

Personnel:	70
Equipment:	5
Barge Charter:	130
Indirect costs:	40

TOTAL COST:	\$245

VIII. PERSONNEL QUALIFICATIONS:

Principal Investigator: Raymond C. Highsmith, Ph.D.

Raymond C. Highsmith is an Associate Professor of Marine Sciences in the Institute of Marine Science, School of Fisheries and Ocean Science of the University of Alaska Fairbanks.

Principal Investigator: Michael S. Stekoll, Ph.D.

Michael S. Stekoll is an Associate Professor with the University of Alaska, Southeast with a joint appointment with the School of Fisheries and Ocean Sciences, University of Alaska, Fairbanks. He has had extensive experience in conducting marine macrophyte research along the coast of Alaska, including research on intertidal *Fucus* populations in Bristol Bay, subtidal and intertidal marine macrophytes around St. Lawrence Island, and kelp research in SE Alaska. He also has published in the area of the effects of oil pollution on marine organisms in Alaska.

Project Leader: Anthony J. Hooten

Andy Hooten is currently enrolled in the doctoral program of biology at George Mason University in Fairfax, Va. He received his master of science degree in zoology from the University of Georgia in 1985. Mr. Hooten served as project leader aboard the Herring Bay experimental field station during the 1990 season. Prior to affiliation with the Coastal Habitat Program, he served as an ecologist for the State of Alaska, Department of Environmental Conservation Oil Spill Response Center.

Project Leader: Peter van Tamelen

Peter van Tamelen is nearing completion of the Ph.D. degree in Zoology from Oregon State University under the direction of Bruce Menge. He has had extensive experience in research in intertidal ecology especially with respect to algal zonation and succession. He served as the project leader for the Herring Bay algal studies during the 1990 field season.

IX. REFERENCES:

- Dayton, P. K. 1971. Competition, disturbance and community organization: the provision and subsequent utilization of space in a rocky intertidal community. Ecol. Monogr. 41:351-389.
- Knight-Jones, E. W. 1953. Laboratory experiments on gregariousness during setting in Balanus balanoides and other barnacles. J. Exp. Biol. 1953 30: 584-598.
- Mann, K.H. and R.B. Clark. 1978. Long-term effects of oil spills on marine intertidal communities. J. Fish. Res. Board Can. (35) 791-795.
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- Southward, A.J. and E.C. Southward. 1978. Recolonization of Cornwall after use of toxic dispersants to clean up the Torrey Canyon spill. J. Fish. Res. Board Can. 35: 682-706.
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X. OTHER INFORMATION:

Refer to Appendices 1 & 2 for the 1990 and 1991 Standard Operating Procedures.

APPENDIX 1

**1990 Invertebrate Standard Operating Procedures
Herring Bay Experimental Field Station**

COASTAL HABITAT INJURY ASSESSMENT: 1990 Standard Operating Procedures for Intertidal Invertebrate Field Experiments in Herring Bay, Prince William Sound, Alaska

The experiments described in these Standard Operating Procedures (SOPs) are an addition to the Injury Assessment process. The presence of a base camp within Herring Bay allows repeated access to study sites to test specific hypotheses which pertain to possible ecological impacts from the Exxon Valdez Oil Spill. Herring Bay was selected as the general location to conduct these studies, due to the heavy levels of oiling experienced, the protected nature of the bay, which provides opportunities for locating non-oiled "control" sites, and the potential use of two "set-aside" sites, which have been left untreated for long term studies.

The selection of study sites is the single most important step for these experiments. Of all the habitat types defined in Prince William Sound, these experiments will be performed in Sheltered Rocky and Sheltered Coarse Gravel environments. These habitat types were selected largely because they represent the most common habitats within Prince William Sound.

In selecting sites for experiments, a range of potential site combinations include the following conditions:

- A. A set aside site;
- B. A non-oiled "control" site;
- C. A mechanically treated site;
- D. A bioremediated site.

A "set aside" is defined as a site that did not receive treatment from Exxon's cleanup effort. "Control sites", are sites which are either truly non-oiled, or have received such a light degree of oiling, that they presently can be determined as "cleaned". "Mechanically treated" sites are defined as those areas which received one or a combination of hand wiping and washing with various water temperatures and pressures. Finally, "bioremediated sites" are those areas which have received an application of Inipol, the chemical used by Exxon to accelerate bacterial degradation of oil.

Based on the above combinations, the original matrix created for site selection appears as follows:

Set Aside	Control	Oiled Mechanical	Oiled Bioremediated.
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This matrix represents sites within one habitat type and serves as one unit for applying experimental treatments. Therefore, replication for any experiment conducted in Herring Bay requires selection of four additional sites to contain the same treatments. Ideally, the matrix listed above would be replicated three times for all experiments in sheltered rocky

habitat types and twice in coarse gravel habitats.

However, after surveys of sites in Herring Bay, and review of data from the Exxon/Federal/State spring shoreline assessments, it is evident that division of sites within the above categories cannot be clearly determined. Virtually all of the segments identified by Exxon and ADEC in Herring Bay have received bioremediation treatments, in addition to any mechanical cleaning, or will have bioremediation occurring during the 1990 treatment season. Furthermore, only one set-aside site of the sheltered rocky habitat type is located in Herring Bay and cannot be replicated within the general vicinity of northern Knight Island.

Therefore, the matrix of site selection can only be stated as impacted versus non-impacted. This results in use of a dual matrix which can be replicated several times, but site selection cannot differentiate between mechanically treated and bioremediated conditions. Thus, for application of the studies presented in this SOP, the modified matrix appears as follows:

IMPACTED	NON-IMPACTED
Oiled	Non-oiled
Mech. Cleaned	"Control" sites

Site Selection.

Careful attention will be given to matching pairs of sites, such that the only differences lie in their oiled and non-oiled conditions. This pairing will include similarity in substrate composition, slope, directional and solar aspect, wave exposure, and common biological communities. In considering pairs of sites to be selected, the following procedures will be conducted and information recorded:

1. Each site considered will be verified by Latitude/Longitude coordinates and compass bearings;
2. The length of the site will be measured at the MHHW line (observed at the base of the Verrucaria zone);
3. The substrate character of the site will be defined (rocky, boulder, coarse grained or a combination of these) and their relative percentages of each;
4. The solar aspect of the site will be determined by compass bearing and recorded;
5. The wave energy/exposure of the site (H,M,L) will be observed and recorded;
6. A detailed drawing of each site will be made; and will include the above factors and major life zones;
7. A video and still photographs will be taken of the site (Attachment I).

Site Characterizations.

In addition to the general description and mapping of each study site, information on the percent cover of major algal and invertebrate species, as well as presence-absence of all intertidal species will provide a basic understanding of community structure and species abundance and distribution (Fig. 1). This information will contribute to a better site description and comparison between study sites.

Each study site will be characterized using the transect and quadrat SOPs of the Coastal Standard Operating Procedure Number 2, and Intertidal Standard Operating Procedure Number 1 (ISOP), with the exception of the differences contained in the following procedures:

1. Because of available working time and the protected nature of Herring Bay, the 35 degree criterion for site workability is rejected, because some of the sites selected can be set up and surveyed by boat (ISOP #1, pg 20). Given the steep nature of these slopes in excess of 35 degrees, the quadrat cannot be allowed to "rest naturally on the substrate". Rather, the quadrat will be allowed to "fit the frame to the slope", but will have to be held in place until the quadrat can be permanently marked with the rotohammer.

2. Each site will be measured based upon obvious traits in site composition, such as changes in substrate character or shoreline topography, or breaks/discontinuity of the intertidal zones of interest. In general, sites will range in length from 15 to 100 M. Each site will have the start and finish of measurement marked with a "medallion" of marine epoxy placed at the base of the *Verrucaria* Zone. Measurement of the site will occur along the edge of the supratidal, at the base of the *Verrucaria* zone. Care will be taken to avoid impacts to the study site; staff will traverse the site in the supratidal zone and in the walking pathway defined for each transect.

3. Note: These rules apply to workable beach lengths, i.e., subtract the beach length of unworkable sections (other habitat types) present within the habitat type from the total beach length of the habitat type before applying these rules.

After the length of the site is determined, divide the total workable beach length of a habitat by the number of transects to be established in the habitat (3 transects are divided into this total length). Call this number X. Essentially, this divides the habitat into intervals of equal length X; one transect will be located in each

interval. Multiply X by a random number. Call this number Y. The first transect will be located Y meters from the left end of the habitat (when facing the beach from the sea). If 3 transects are to be established in the habitat, they should be positioned X meters and 2X respectively, to the right of the first transect.

4. Quadrat size is 80x50 cm. Collect data for the presence/absence of all species;

5. Collect data for percent cover of sessile invertebrates and algae. Follow ISOP #1; remove all drift *Fucus* from both 40 X 50 cm areas. Remove the *Fucus* canopy from the right half of the left 40 X 50 cm quadrat. Photograph both halves per ISOP #1, 1990 Quadrats.

6. A swath survey for macro invertebrates will be conducted pursuant to ISOP # 2.

7. With item H of ISOP #1 (limpet/mussels/*Fucus* semicircle, only limpet data will be collected at this site. Aside from the biological data collection specified here, all other biological collections identified in ISOP #1 are ignored.

8. Collect Hydrocarbon data pursuant to Coastal Habitat SOP #4.

8. Move to the next quadrat and repeat steps 4-7.

These transects will be used for site characterization for presence/absence of all species, and percent cover of macro algae, barnacles and mussels. Figure 2 shows the layout of sampling procedures for site characterization.

Transplanting of oiled and non-oiled substrates.

The objectives of this experiment are to examine differences in: a) the percent cover of barnacles and macro algae; b) the number of individuals per unit area; and c) the presence/absence of species on oiled and non-oiled substrates placed within the various sites identified.

Seventy two oiled rocks are to be collected from a heavily oiled site within Herring Bay. These rocks will be of the same size and geological type and will represent substrate coated with 1 year old Exxon Valdez Prudhoe Bay Crude (EV). Also, 72 rocks of the same size and geological type will be collected from the same or a geologically identical, but unoiled beach. All rocks collected are to be packed in boxes or coolers and separated by aluminum foil so that the rocks do not touch one another.

Upon return to the laboratory, all rocks will be laid out and one half of each rock will be cleaned with Methylene Chloride (MeCl₂), with the exception of 8. These 8 will be left totally oiled to serve as strata for samples of oil weathering analysis. Also, 8 of the non-oiled rocks will have their entire surface cleaned with MeCl₂ to serve as controls for use of the solvent. Care should be taken when handling this solvent. Solvent resistant gloves should be worn at all times and work should be conducted in a well ventilated area. Cleaning of each rock will involve either dipping one half of the rock in several baths of MeCl₂, or hand-wiping or both. After this half is thoroughly cleaned, the rocks will be allowed to dry. "Top" and "bottom" of each rock will be determined with regard to symmetry and morphology. The "top" will be assigned to the surface with the least irregularity. When dry, each rock will receive a unique identification number with an indelible marker. Each rock will then be measured with calipers for total length, and the length of the cleaned sides. These data will be recorded on a data form (Fig. 3). Each rock will also be photographed and the frame number will be logged on the data form.

For the non-oiled rocks collected, half of each of these rocks surfaces will be dipped with fresh Prudhoe Bay Crude (PBC) until a "tarred" coating is achieved. These rocks will be allowed to dry. Again, each of these freshly oiled rocks will be measured for total length and clean length and photographed.

In addition to the rocks, a total of 72 clay tiles will also be incorporated into the experiment to be placed among the rocks. Half of these (36) will be oiled with fresh PBC and the other half will remain clean. These tiles will be placed in the field in oiled and unoiled pairs.

At each of the experimental sites, rocks and tiles will be placed at the 2 m elevational contour. Each site will receive an identical number of rocks and tiles representing the following experimental conditions (Fig. 4):

The basic experimental unit is six rocks and six pairs of tiles. Three of the rocks are from the original oiling by EV and cleaned with MeCl₂. The remaining three rocks were one-half treated with fresh PBC. A pair of clay tiles consists of one tile treated with fresh PBC and one unoiled tile. In addition to this basic experimental unit, there are 3 sets of 3 rocks representing EV and 3 sets of 3 rocks representing fresh PBC. These are placed in the field as destructive samples, with each set to be retrieved at three separate time periods. These time periods are mid summer 1990, early fall 1990, and mid spring, 1991. Finally, at each site, one completely oiled rock with EV and one with fresh PBC and one entirely cleaned rock

with MeCl_2 , will serve as sources for weathering analysis and cleaning control for MeCl_2 , respectively. Refer to Attachment 2 for the SOP for oil weathering analysis.

For the purposes of this SOP, each of these experimental units (33 total per site) is referred to as a "batch" of rocks and tiles. This "batch" will represent all substrate combinations to be located at one site. Using a random numbers generator on a calculator, each uniquely identified rock (i.e. those freshly painted with PBC and last year's coating with EV) will be assigned to a "Batch". These batches of rocks and tiles will then correspond to numbered poker chips, and each chip will be drawn and assigned to the matrix of study sites. Each batch of rocks and tiles will be boxed and taken to their respective sites.

Once in the field, a uniform substrate at the 2 meter elevation contour of each site will be chosen, video taped and mapped. Establishment of the second meter of vertical fall is pursuant to IOP#1.

The marine epoxy used to glue the rocks to the rock substrate is a 1:1 mixture of a two-part compound. Working time of the epoxy is approximately 30 minutes.

It is important to ensure that all rocks and tiles are placed along the same elevation contour. Furthermore, the substrate should be scraped free of most barnacles and macro algae to ensure a firm setting of the epoxy to the substrate. Once the specific location is established, poker chips will be placed into a bag, shaken and drawn to determine whether an EV or PBC, tile pair, or control rock will be placed first. After this determination, the placement of these units will be alternated based upon the order drawn, until each batch is completely laid out. Obviously, those units with lessor replication will be exhausted first, eventually resulting in alternation only between EV and PBC rocks. This procedure is to be repeated at each site.

After placement, each site will be visited once a week during low tide series. Observations as to settlement by barnacles and macro algae on rock surface and surrounding settlement conditions will be recorded. Also, individual species will be identified, counted and recorded. Counting will involve use of a 3 cm X 3 cm quadrat. The quadrat edge is placed at the midpoint of the line separating the oiled and unoled portions of the rock. Rocks will be photographed at a fixed

focal length to incorporate a 5 cm X 10 cm area. From left to right along the site, the first 3 rocks of EV, the first 3 rocks of fresh PBC, the clay tiles, and all control rocks are to remain affixed indefinitely, and will be examined in the field and through macro photography. However, the remaining rocks are to be retrieved from left to right at 3 separate time series. These time series will occur in mid-summer, 1990, early fall 1990, and spring of 1991. After field examination and photography, these rocks will be returned to the laboratory and organisms that settled will be counted. Counting will involve either a point method for settlement or a magnified transect and micro photographing of the rock surface. Also, scraping of the rock surface and preparation of slides for micro algae and diatom identification may be attempted.

Limpet Grazing

At each of eight sites, algal beds will be identified and measured at two tidal contours (i.e. meters of vertical fall). Because treatment of oiled sites by Exxon resulted in removal of the upper elevational zones (particularly *Fucus*), the first elevational contour is defined at the control sites. Fences will be placed at the first meter of vertical fall contour within the *Fucus* zone based. As an example, if the *Fucus* begins at MHHW, then the transect is placed at the 1m contour. This procedure will be replicated at the contour of the impacted shoreline, where mechanical treatment such as high pressure-hot water washing, may have removed large concentrations of *Fucus* cover. This first contour at the impacted sites will also be verified by evidence of *Fucus* holdfasts (plant bases) or stipes, occurring within the contour.

The objective for the second elevational contour was to examine grazing in an algal dominated zone other than *Fucus* (i.e. *Cladophora*). The oil-impacted sites are used to determine placement of fences. Because Exxon was instructed to treat the shorelines down to only the mid-intertidal zone, all fences will be established at the beginning of this *Cladophora* zone, where impacts from treatment activities can be observed. The elevational contour of this observed zone is averaged within the site, and all fences are placed along this selected contour, regardless of the variation of algal bed density throughout the contour level. For example, if *Cladophora* zone and observed impacts begin at 2.25 m, then the location of fences will be established in this zone. This procedure dictates the second contour at control sites. Consequently, the second contour level may vary between pairs, but is identical at each control and impacted pair.

Once the algal beds are measured, subtract 0.25 meters on either side to ensure the sample area is buffered. Each location should preferably be homogenous in substrate composition, such as an algal bed on a rock outcrop.

Eight fences will serve as a base unit. With the 2 tidal heights, this will constitute 16 fences per site (Fig. 5) Fences will be structured within the substrate by marine epoxy and hard ware cloth. Fences will be 25 X 25 cm in area, and will be randomly placed within each site. The length of each algal bed is measured and the appropriate buffer subtracted. Eight fences are divided into the resulting length, giving a segment length, and this number is multiplied by a random value. The resulting value establishes location of the first transect. Subsequent fences are placed one segment length from the previous fence. Each fence is placed at the transect mark at the selected elevation contour. Place the fence at this contour with the upper left corner of the fence serving as the marked point. As an example, a 25 meter algal bed minus 0.25 meters on each side equals a 24.5 meter workable area. Eight fences divided into this length equals 3 meters per fence. Three times the random value 0.98 equals 2.94 meters for the first fence. Therefore, the 2nd - 8th fences are placed every 3 meters after 2.94 m. Should a fence location be determined as unworkable, a rule is established to move the location 25 cm to the left until the first workable area is encountered. "Unworkable" is defined as an area that is not uniform for placement of a 25 X 25 cm fence.

Use of marine epoxy for anchoring the fences will require cleaning small strips within the algal beds to ensure proper adhesion of the epoxy. This should be done with as little disturbance to each site as possible so that the interior to each fence still retains all of its original algal cover. After the epoxy begins to set, place the fencing into the epoxy base and tie the corners with wire. Also, construct a small lip to prevent escape and entry of limpets over the fence.

After these fences are established at each site, large numbers of limpets from the genera *Lottia* and *Tectura* will be collected at a site away from all study areas. No individuals less than 10 mm in shell length will be used for this study, based upon difficulty in differentiation among genera below this size. The selected individuals will be measured, weighed and sorted in to genus and size classes, and uniquely identified and marked. The method of marking will involve a miniature tag (i.e. electronic wiring number labels & epoxy) glued to the shell of the limpet.

Based upon an average density of limpets determined by transects, a representative limpet from each genera and size class will be selected to form a "batch" of limpets (X). To

exemplify, if the transects disclose a mean density of six limpets per unit area, then six limpets of representative size and species distribution constitutes a single "batch" of limpets. These "batches" will serve as the base unit for placing numbers of limpets in fences. Each batch will be placed into customized containers and kept in fresh seawater for holding.

A randomized selection of fence treatments will occur by use of poker chips representing the cells of the following experimental matrix:

	2X	X	X/2	0
ALGAE				
NO ALGAE				

This process will be re-randomized at each site.

The batches of limpets will be numbered, and poker chips will serve as representatives for assigning groups to the fences at each site. All batch-numbered poker chips will be placed in a bag and drawn randomly to be assigned to each of the fences and treatments. Batches will be assigned according to the above matrix. For the X/2 treatments, one batch of limpets will be divided between two fences.

For those fences selected for algal removal, the entire 625 cm area within the fences will have all algae, herbivores and limpet predators removed. The algae will be bagged and returned to the laboratory for wet and dry weight measure. With the remaining cages, all mobile invertebrates (i.e. *Littorina*, *Nucella*) will be removed, and total percent algal cover, as well as percent cover by algal species will be estimated and determined by a point grid method. Also, a photograph will be taken of each fenced area.

Limpets will then be placed in each fence according to their batch assignments. The fences will be revisited during low tide periods on a weekly basis. Numbers of surviving individuals will be recorded; dead limpets will be removed and their shells measured (Fig. 6). Also, new recruits will be recorded, but removed. The total percent algal cover and percent cover by algal species will again be determined by a point grid method.

This experiment will be run throughout the summer, or until all limpets have died. Survivors will be remeasured and reweighed. At the termination of the experiment, all algae will be removed and bagged, identified in the laboratory, and a final wet and dry weight will be measured. When the experiment is terminated, all fence material will be removed from each site.

Population Dynamics of Limpets, *Littorina*, *Nucella* and *Leptasterias hexactis*.

The objectives of this study are to monitor differences in numbers of limpets, *Littorina*, *Nucella*, and *Leptasterias hexactis*, between impacted and non-impacted sites.

Transect heads will be established pursuant to Coastal Habitat operating procedures number 1 (Fig 7). The length of each site will be divided by 6 and multiplied by a random number to locate transect heads. Quadrats will be located randomly along each transect as in Site Characterizations. Permanent quadrats will be established within the first, second and third meters of vertical fall along each of these six transects. Use of a rotohammer will mark the upper left and bottom right of each quadrat.

Quadrat dimensions are 20X50 cm. Once in place, count all limpets, *Littorina*, *Nucella*, and *Leptasterias hexactis* within the quadrat and record this data on the data form (Fig. 8).

Measure a 1 m semicircle adjacent to and centered at the left of the 20X50 cm quadrat. Measure the distance from the center of the base of the semicircle to the closest limpet, *Littorina*, *Nucella*, and *Leptasterias hexactis* or 1.0 m if none of these species are found in the semicircle.

ATTACHMENT 1

Videography of Sites

Each site selected will be recorded on videotape during a low tide period. This record will serve as an aid to site description.

Each "Control" site and its oiled counterpart is filmed. Panning of each site from a skiff will occur from left to right. Site number and the experiments being conducted on the site is recorded on the audio portion of the tape.

After the distant filming of a given site, an on-shore filming of the 0 meter mark is taken. The video camera then pans to the right to view as much of the site as possible. The aspect of shooting is from the upper intertidal.

If the site is of significant length (i.e. 50 m or greater, or heterogeneous in shoreline topography) the video camera will record intermittent markings (i.e. z-spar epoxied meter marks) of the site, while panning to the right of the mark, and then back to the left to show reference to the beginning of the site.

Finally, the end of the site is videotaped, showing the end site mark, with total meters and site number marked in an epoxied "medallion" placed at the base of the Verrucaria zone. The video camera then pans across the entire site, back toward the zero meter mark.

ATTACHMENT 2

Settlement on Oiled and Un-oiled Substrates: Oil Extraction for "Weathering" Analysis.

Throughout this substrate transplant experiment, the chemical composition of crude oil will change over time. Consequently, the "thickness" of oil coating of the substrates will gradually decrease. The purpose of this procedure is to quantify a rate of change in the oil's character. This procedure employs a simple gravimetric analysis of an area of oil, extracted by Methylene Chloride (MeCl₂).

Several of the substrates placed in the field are solely for the purpose of these extractions. On a monthly basis, a 3 cm² area of both an EV rock and PBC rock will be wiped with a methylene chloride saturated wipe (i.e. kimwipe). This wipe will be pre-weighed and placed in a small vial containing 25 ml of MeCl₂.

After wiping, the wipe will be returned to the laboratory. All MeCl₂ will be allowed to evaporate, and the wipe will be reweighed and re-stored in the same vial with an additional 25 ml of MeCl₂. This vial will be refrigerated for possible Gas Chromatography/Flame Ionization Detection (GC/FID) analysis.

FIGURE 1.

Form Used in Recording Site Characterization Data

CHIA/JAF HERRING BAY EXPERIMENTAL STATION

Site Characterization Log Sheet

Date: _____ Site Number: _____

Time: _____ Site Name: _____

Weather: sunny partly cloudy cloudy rain snow

(circle)

Samplers: M. Derenoff G. Hollowell A. Hooten T. Lewis

G. Reedy F. Roddy C. Sullivan P. Van Tamelen _____

(circle)

Transect Number: _____

Quadrat # _____

Procedural Checklist

___ photograph left 40x50 area with drift algae removed (_____ roll and frame #)

___ remove Fucus from right 40x50 cm area

___ photograph right quadrat (_____ roll and frame#)

___ determine total percent cover _____%(left 20x50)

_____%(right 20x50)

___ complete species checklist on reverse of this page

___ measure nearest limpet within 1 meter: _____ (enter 1m if none)

___ collect five nearest limpets (see reverse)

Quadrat # _____

___ photograph left 40x50 area with drift algae removed (_____ roll and frame #)

___ remove Fucus from right 40x50 cm area

___ photograph right quadrat (_____ roll and frame#)

___ determine total percent cover _____%(left 20x50)

_____%(right 20x50)

___ complete species checklist on reverse of this page

___ measure nearest limpet within 1 meter: _____ (enter 1m if none)

___ collect five nearest limpets (see reverse)

Quadrat # _____

___ photograph left 40x50 cm area with drift algae removed (_____ roll and frame #)

___ remove Fucus from right 40x50 cm area

___ photograph right quadrat (_____ roll and frame#)

___ determine total percent cover _____%(left 20x50)

_____%(right 20x50)

___ complete checklist on reverse of this page

___ measure nearest limpet within 1 meter: _____ (enter 1m if none)

___ collect five nearest limpets (see reverse)

SPECIES CHECKLIST

<u>ALGAE</u>	<u>Q1</u>	<u>Q2</u>	<u>Q3</u>	<u>INVERTEBRATES</u>	<u>Q1</u>	<u>Q2</u>	<u>Q3</u>
Acrosiphonia arcta	___	___	___	Anthropleura artemisia	___	___	___
Audinella purpurea	___	___	___	Balanus glandula	___	___	___
Blidingia chaudefaudii	___	___	___	Chthalamus dalli	___	___	___
Cladophora sericea	___	___	___	Halobisium accidentale	___	___	___
Cryptosiphonia woodi	___	___	___	Kathanna sp.	___	___	___
Devaleraea ramentacea	___	___	___	Ligia sp.	___	___	___
Dumontia contorta	___	___	___	Littorina scutulata	___	___	___
Endocladia muricata	___	___	___	Littorina sitkana	___	___	___
Enteromorpha linza	___	___	___	Lottia borealis	___	___	___
Enteromorpha intestinalis	___	___	___	Lottia pelta	___	___	___
Fucus gardneri	___	___	___	Lottia sp.	___	___	___
Fucus sp.	___	___	___	Macoma balthica	___	___	___
Gloiopeltis furcata	___	___	___	Mytilus edulis	___	___	___
Halosaccion glandiforme	___	___	___	Modiolus rectus	___	___	___
Hildenbrandia rubra	___	___	___	Nereis sp.	___	___	___
Mastocarpus papillatus	___	___	___	Nucella emarginata	___	___	___
Myelophycus intestinalis	___	___	___	Nucella lamellosa	___	___	___
Neorhodomela aculeata	___	___	___	Polychaeta sp.	___	___	___
Ondontalia floccosa	___	___	___	Searlesia dira	___	___	___
Phycodrys rigii	___	___	___	Semibalanous balanoides	___	___	___
Pilayella washingtoniensis	___	___	___	Semibalanus cariosus	___	___	___
Polysiphonia senticulosa	___	___	___	Siphonaria thersites	___	___	___
Prasiola borealis	___	___	___	Spirorbis sp.	___	___	___
Ptilota pectinata	___	___	___	_____	___	___	___
Ralfsia fungiformis	___	___	___	_____	___	___	___
Rhodomela subfusca	___	___	___	_____	___	___	___
Scytosiphon lomentaria	___	___	___	_____	___	___	___
Ulothrix implexa	___	___	___				
Ulva fenestra	___	___	___				
Yendonia crassifolia	___	___	___				
Corallines: articulated ___ encrusting ___							

limpet data from reverse

<u>Q3</u>	<u>species</u>	<u>length</u>	<u>weight</u>
1.			
2.			
3.			
4.			
5.			

<u>Q1</u>	<u>species</u>	<u>length</u>	<u>weight</u>
1.			
2.			
3.			
4.			
5.			

Comments:

<u>Q2</u>	<u>species</u>	<u>length</u>	<u>weight</u>
1.			
2.			
3.			
4.			
5.			

Signature: _____

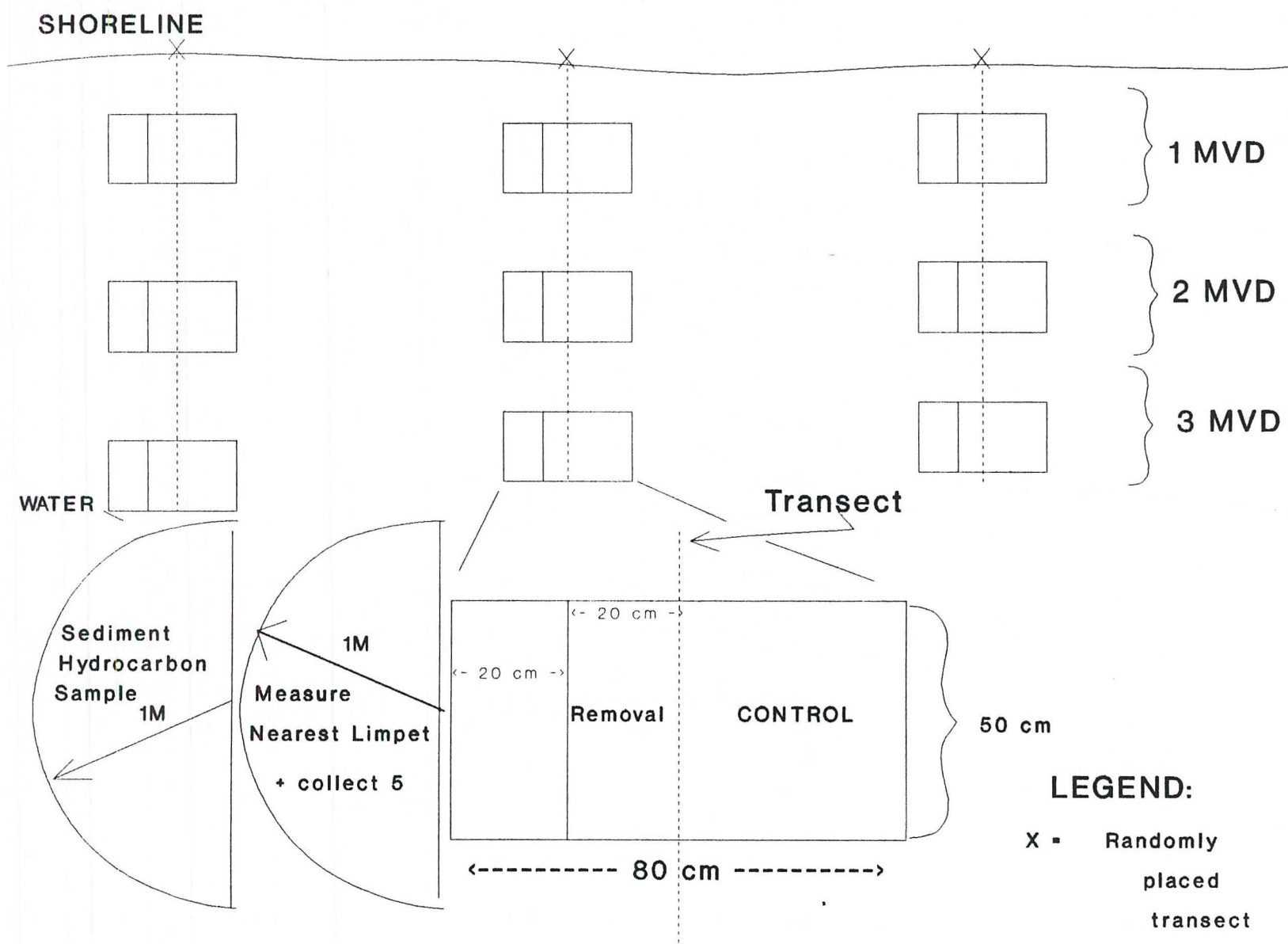


Figure 2. Placement of quadrats for Site Characterizations

FIGURE 3.

Form Used for Recording Settlement Data on Oiled and Non-Oiled Substrates.

CHIA/UAF HERRING BAY EXPERIMENTAL STATION

R X Δ Log Sheet

Date: 11 July 90

Site Number: 1723 X

Time: 1017

Site Name: Barnacle Pt

Weather: sunny partly cloudy (circle) cloudy rain snow

Samplers: M. Derenoff G. Hollowell A. Hooten T. Lewis G. Reedy
 F. Roddy C. Sullivan (circle) P. Van Tamelen

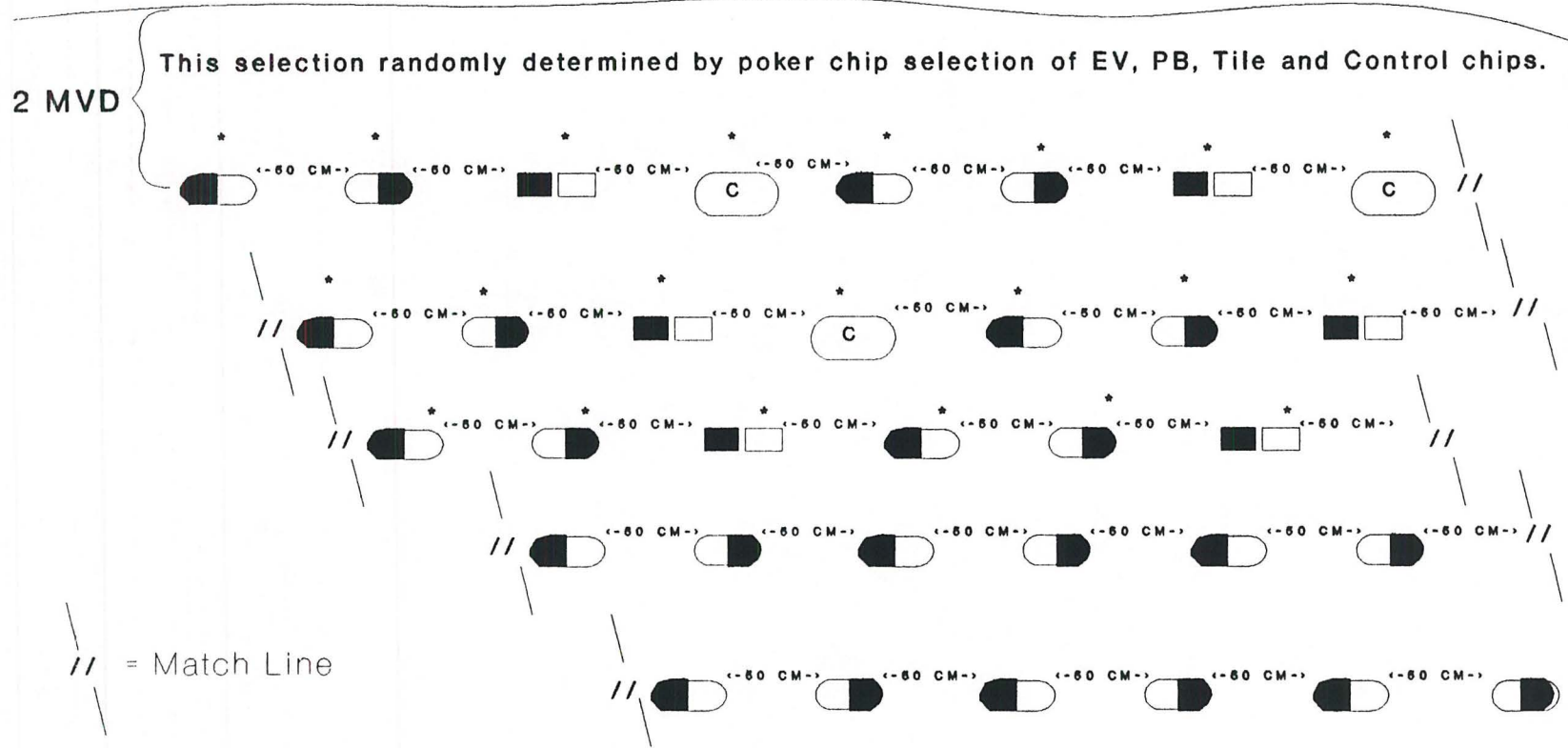
Legend: Unit #'s EV = 1+ year old Exxon Valdez crude oil
 PB = fresh Prudhoe Bay crude oil
 T = tile pair (oiled vs. non-oiled)

** NOTE: Report all data as O/U for oiled vs. non-oiled substrate.**

Counts done in 33m² subsection of units.

Unit Number	New Settlement? O/U (y/n)	# Barnacle Juveniles	# Fucus Germlings	Other (define)	Roll and Frame #	Pics O. Ch.
1. T ₁	n/n	0			X: 24 (cont.) 10, 11, 12	
2. PB ₁₂	n/n	0			13, 14, 15	
3. PBow					16	
4. EV ₆₀	y/y	3/7			17, 18, 19	
5. T ₂	n/n	0			20, 21, 22, 23	
6. PB ₆₅	y/y	80/38			X: 25 1, 2, 3	130ft
7. C1723X M	y/y	254/269			4, 5, 6	
8. EV ₁₉	y/y	25/32			7, 8, 9	
9. T ₃	y/y	3/18			10 Non oiled	
10. PB ₂₀	y/y	171/250			11, 12, 13	
11. C1723X ELOW					14	
12. EV12	y/y	103/37			15, 16, 17	
13. T ₄	y/y	4/3			18 Non-oiled	
14. PB55	y/y	3/6			19, 20, 21	
15. EV45	y/y	223/331			22, 23, 24	

SHORELINE



LEGEND:





 Rock w/ 1 yr Exxon Valdez oil (EV)	 Rock w/control for Methylene Chloride or oil weathering analysis for EV or PB
 Rock w/ fresh Prudhoe Bay Crude (PB)	
 one pair tiles, oiled and non-oiled w/ PB	* = Basic experimental unit, placed in field indefinitely. Remaining EV & PB rocks destructively sampled during three separate dates- summer & fall, 1990, spring, 1991.

Figure 4. Placement of oiled and non-oiled substrates.

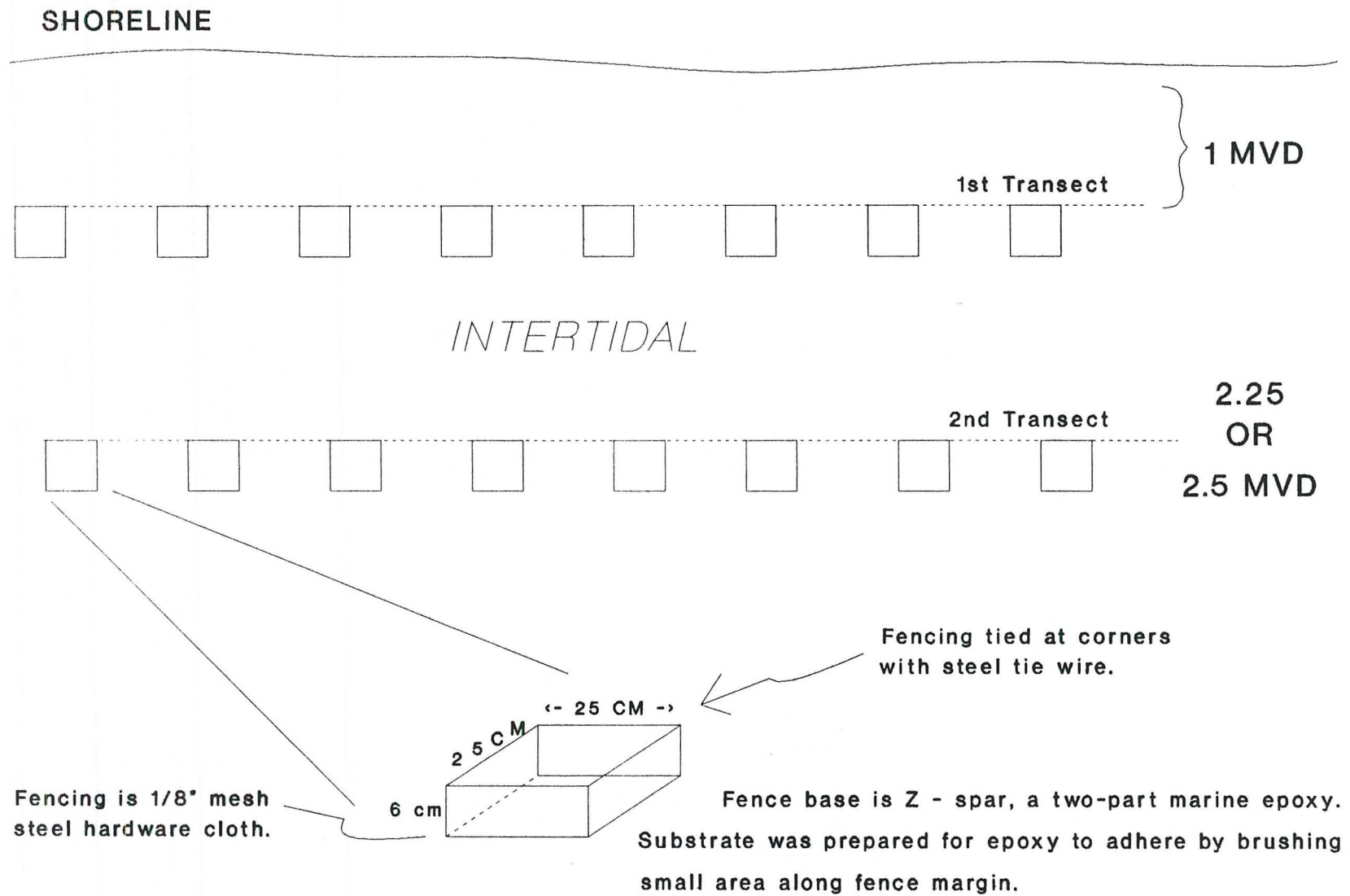


Figure 5. Placement of fences for Limpet Grazing Experiment.

FIGURE 6.

Form Used for Recording Limpet Grazing Data

CHIA/UAH HERRING BAY EXPERIMENTAL STATION
Limpet Grazing Log Sheet

Date: 7-13 Time: 1100 Site Name: Waikiki

Contour: 2.0 MVD Site Number: 1311X

Weather: sunny partly cloudy (circle) cloudy rain snow

Samplers: M. Derenoff G. Hollowell A. Hooten T. Lewis G. Reedy
F. Roddy C. Sullivan P. Van Tamelen (circle)

Comments:

number remaining last time

<p>1 AX Limpets remaining (5) 326 ①</p>	<p>Algae present 1. F.c.y 2. Cd.s 3. Mp.i. 4. Rd.s 5. Palmaria 6. Hb.c. 7. 8.</p>	<p>Estimated % cover (10 + 15) .5 = 12.5 % (5 + 10) .5 = 7.5 % (3 + 3) .5 = 3 % (3 + 7) .5 = 5 % (1 + 1) .5 = 1 % (1 + 2) .5 = 1.5 % (+) .5 = % (+) .5 = %</p>	<p># dots covered 17 / 100 = 17 % 10 / 100 = 10 % 3 / 100 = 3 % 11 / 100 = 11 % 1 / 100 = 1 % 5 / 100 = 5 % / = % / = %</p>	<p>Total Algal Cover: 42 / 100 = 42 % comments: 1 L.p removed 5 overlapping</p>
<p>2 2X Limpets remaining (14) 192 232 505 282 562 506 425 240 230 644 149 547 ②</p>	<p>Algae present 1. \emptyset 2. \emptyset 3. 4. 5. 6. 7. 8.</p>	<p>Estimated % cover (+) .5 = % (+) .5 = % (+) .5 = % (+) .5 = % (+) .5 = % (+) .5 = % (+) .5 = % (+) .5 = %</p>	<p># dots covered / = % / = % / = % / = % / = % / = % / = % / = %</p>	<p>Total Algal Cover: / = % comments: removed 1 L.p. removed 1 N.I.</p>
<p>3 \emptyset Limpets remaining</p>	<p>Algae present 1. Hb.c. 2. 3. 4. 5. 6. 7. 8.</p>	<p>Estimated % cover (2 + 1) .5 = 1.5 % (+) .5 = % (+) .5 = % (+) .5 = % (+) .5 = % (+) .5 = % (+) .5 = % (+) .5 = %</p>	<p># dots covered \emptyset / 198 = % / = % / = % / = % / = % / = % / = % / = %</p>	<p>Total Algal Cover: \emptyset / 198 = 0 % comments: removed 1 N.I. removed 1 sea star</p>
<p>4 X/2 Limpets remaining (1) \emptyset</p>	<p>Algae present 1. Hb.c. 2. Cd.s. 3. 4. 5. 6. 7. 8.</p>	<p>Estimated % cover (1 + 3) .5 = 2 % (1 + 1) .5 = 1 % (+) .5 = % (+) .5 = % (+) .5 = % (+) .5 = % (+) .5 = % (+) .5 = %</p>	<p># dots covered 4 / 100 = 4 % \emptyset / 100 = 0 % / = % / = % / = % / = % / = % / = %</p>	<p>Total Algal Cover: 4 / 100 = 4 % comments:</p>

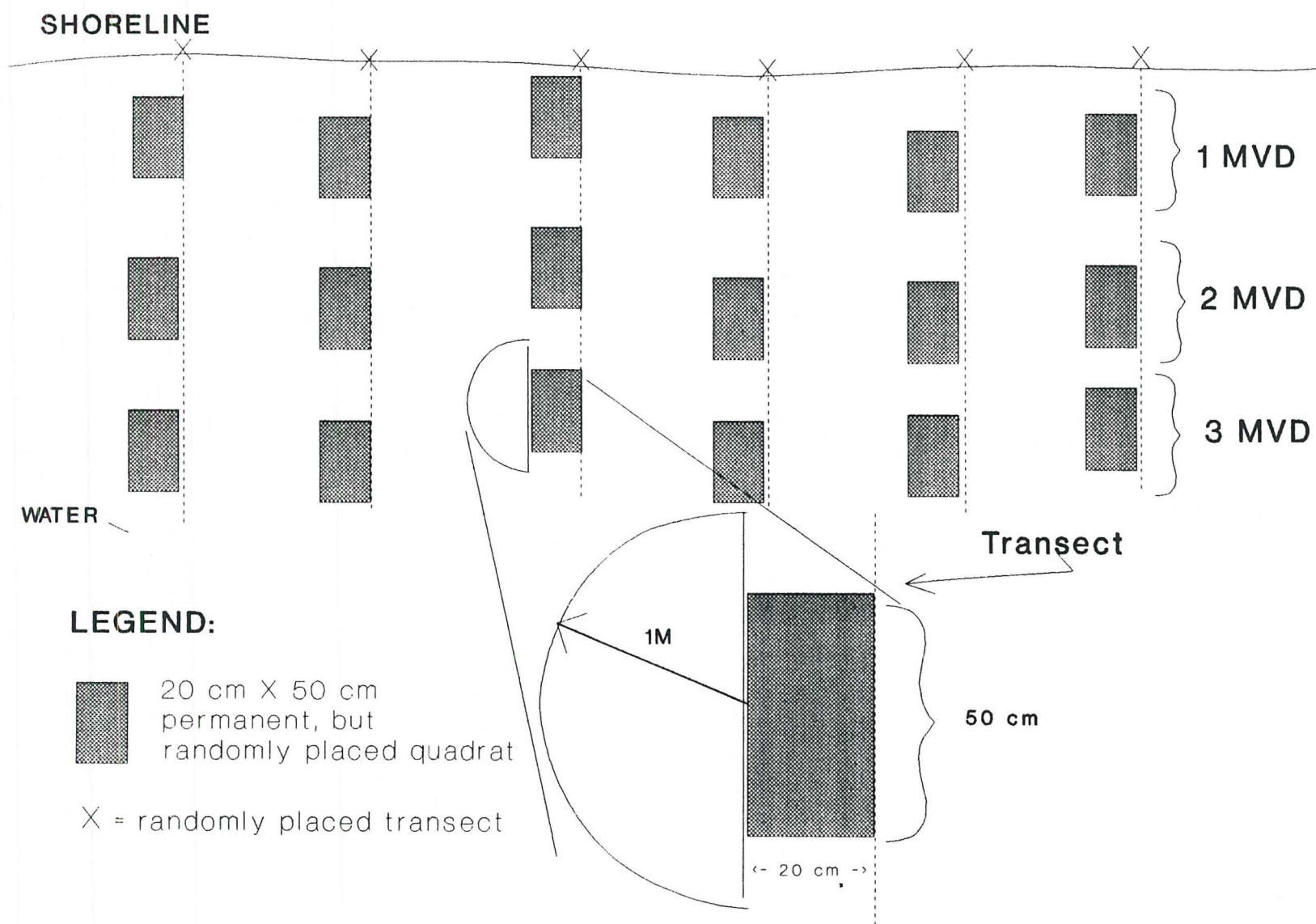


Figure 7. Placement of Permanent Quadrats for Population Dynamics of Limpets, Littorina and Nucella

FIGURE 8.

Form Used for Recording Invertebrate Population Dynamics Data

CHIA/UAH HERRING BAY EXPERIMENTAL STATION
Invertebrate Population Dynamics

*Control
MJD
10/20*

Site Name: ANCHOR BEACH
Site Number: 2834X

Date: 08/22/90 Signature: MJD
Time: 8:08 am (comments on reverse)

Weather: sunny partly cloudy cloudy rain snow
(circle)

Samplers: M. Derenoff G. Hollowell A. Hooten T. Lewis G. Reedy
C. Sullivan F. Roddy P. Van Tamelen

(circle)

		# of Lottia	# of T. persona	# of T. scutem	# of unknown limpets	Σ of all limpets	# of N. emarginata	# of N. lamellosa	Σ of all Nucella	# of L. sitkana	# of L. hexactis	distance to nearest Littorina	distance to nearest limpet (L., T.P. or T.S.) (UNK.)	distance to nearest Nucella (N.R. or N.I.)
transect 1	1m drop ₁		3			3			0	2	0	4.5cm	19cm ^{T.P.}	>1m
	2m drop ₂	1	11		3	15			0	0	0	>1m	6cm ^{T.P.}	60cm ^{Ne.}
	3m drop ₃		1			1			0	0	0	>1m	38cm ^{L.P.}	>1m
transect 2	1m drop ₄					0			0	14	0	10cm	43cm ^{L.P.}	>1m
	2m drop ₅		3			3			0	8	0	38cm	1cm ^{T.P.}	>1m
	3m drop ₆	6	10		61	77			0	0	0	36cm	4cm ^{T.P.}	94cm ^{Ne.}
transect 3	1m drop ₇					0			0	1	0	26cm	>1m	>1m
	2m drop ₈		2			2			0	4	0	4cm	44cm ^{T.P.}	>1m
	3m drop ₉					0			0	12	0	4cm	29cm ^{T.P.}	>1m
transect 4	1m drop ₁₀					0			0	5	0	7cm	>1m	>1m
	2m drop ₁₁	1	1			2			0	7	0	6cm	35cm ^{L.P.}	>1m
	3m drop ₁₂				1	1			0	20	0	9cm	21cm ^{T.P.}	>1m
transect 5	1m drop ₁₃					0			0	3	0	9.5cm	23cm ^{T.P.}	>1m
	2m drop ₁₄		2			2			0	12	0	3.5cm	7cm ^{T.P.}	>1m
	3m drop ₁₅	4	8		15	27			0	2	0	>1m	14cm ^{T.P.}	4.7cm ^{Ne.}
transect 6	1m drop ₁₆					0			0	1	0	37cm	56cm ^{T.P.}	>1m
	2m drop ₁₇					0			0	13	0	3cm	7cm ^{L.P.}	>1m
	3m drop ₁₈	1			15	16			0	3	0	>1m	22cm ^{UNK.}	43cm ^{Ne.}

#1: # of Pilayella/Cladophora LYING OVER ROCKS.
#2: drift plants covering 10% of a.

APPENDIX 2

**1991 Invertebrate Standard Operating Procedures
Herring Bay Experimental Field Station**

COASTAL HABITAT INJURY ASSESSMENT:
Standard Operating Procedures
Intertidal Invertebrate Field Experiments
Herring Bay, Price William Sound, Alaska: 1991

These 1991 Standard Operating Procedures (SOP) serve as a supplement to the 1990 barge SOP. A copy of the 1990 SOP is appendix 1. Any changes, or exceptions to studies from 1990 are identified herein.

Site Selection:

Additional sites may be selected to increase replication of some of the 1990 studies. The selection of sites will be based upon availability, to effectively match both impacted and control pairs. Any additional site selection in 1991 will be implemented pursuant to the 1990 SOP.

Population Dynamics of Limpets, *Littorina*, *Nucella* and *Leptasterias*.

The population dynamics monitoring as described in the 1990 SOP will be continued at all 10 sites. For 1991, size class determination of each species examined will be included in the normal census. When quadrats are conducted at each study site, Limpets, *Littorina sitkana*, *Nucella* spp. and *Leptasterias hexactis* will not only be counted, but each count will be divided into size classes for each of these species. If possible, additional site pairs for population dynamic studies will be established.

Transplanting of oiled and non-oiled substrates.

With the 1990 experiment, the last time-series sample is to be retrieved in April. The remaining experimental units will be left in the field indefinitely and monitored during the 1991 season. However, as a supplement to this original experiment, new red clay tile pairs (both tarred and cleaned) will be added to each site in a manner identical to the procedure defined in the 1990 SOP. Six pairs of tarred and cleaned tiles will be replaced at each study site, with one pair destructively sampled each month, and quantified for barnacle settlement in the laboratory. At each site, four pairs will be destructively sampled over the course of the field season, and two pairs will remain on site through the duration of the experiment. These tiles will be placed in those identical locations where time series units were removed during 1990. These locations can be easily determined from the permanent units remaining at each site, and will preserve the original random assignment of units. Units remaining in the field will continue to be quantified pursuant to the 1990 SOP.

Also, one additional oiled tile placed at each site will serve as a source for oil weathering analysis. Refer to Attachment 2 of the 1990 SOP for the oil weathering analysis procedures.

Mussel Densities:

As a result of the CHIA intertidal sampling in 1989 and 1990, an increase in the abundance of mussels at oiled sites has become evident. This may be explained by certain species of filamentous algae recolonizing at oiled sites free from grazing pressure, and recruiting juvenile mussels benefitting from these algae, which serve as anchoring substrate. The following experiment tests this hypothesis by clearing small areas and excluding all but algae and mussels and monitoring mussel density over time.

Three pairs of oiled and control sites with evidence of mussel populations will be selected for this experiment. At each study site three transect heads will be randomly selected pursuant to the methods identified for transect selection (1990 SOP). At each of these points, an area at the 1.5 meter contour (from MHHW) will be selected. This area, 0.5 m in radius, will be cleared of all algae and invertebrates. A circular fence of 1/8" mesh steel hardware cloth (10 cm in height) and marine epoxy will be constructed around the boundary of the cleared area.

Each site will be monitored three times each week to ensure that all grazers/predators are removed from the fence. Once each month, a random dot grid will be placed over the fence and a patch of filamentous algae 5 x 5 cm will be selected and removed from each fence. This sample will be placed in a whirlpak bag and returned to the laboratory. Each sample will be analyzed for mussel content, and the total number of mussels in each sample will be counted. Counting will involve use of a dissecting scope and a hand-held counter. Counting of each sample will be repeated until the coefficient of variance of the mean does not exceed 0.05. If too many mussels are present in the sample, then a subsample of the original will be quantified as above.

Site Characterizations:

Site characterizations will remain a lower priority compared to the specific experiments conducted at each of the study sites. Each of the sites originally selected during the 1990 field season will again be characterized pursuant to the 1990 SOP, and will be visited three times during the beginning, middle and end of the field season. However, no new additional sites that may be selected for expansion of any of the detailed experiments will be added to the site characterization process.

Barnacle recolonization:

This experiment was not an original part of the 1990 SOP. Therefore, materials and methods used during 1990 are presented in this SOP, and a discussion to expand this experiment in the 1991 field season follows.

Within Herring Bay, certain oiled locations have heavy accumulations of dried tar, especially in the upper intertidal zone, where desiccation and baking by sunlight have resulted in an asphalt condition of the oil. The purpose of this study was to examine whether the presence of tarred upper intertidal areas reduces the settlement capability of barnacle larvae relative to cleaned areas within a tarred substrate. Further, does the presence of oil reduce the survival of barnacle juveniles, and how do such differences compare to barnacle settlement at reference sites?

Two oiled sites and two reference sites of similar character were selected in Herring Bay for this study in 1990. The oiled sites selected have vertical rock faces where barnacles presently exist, or in the case of sites that were heavily oiled and treated, having skeletons still attached to the substrate. The non-oiled reference sites chosen had high densities of the barnacle *Semibalanus balanoides*.

At each site, paired 10 X 10 cm plots were established. One member of each pair was scraped and brushed to remove all visible tar (or barnacles in the cases of the non-oiled sites). The length of each site was measured, and the number of planned pairs divided into the site length. The first plot was placed randomly, within the first segment, and subsequent plots were placed at equal distances from the first. A coin was flipped to determine which 100 cm² area of the first pair to scrape. The subsequent scraped plots were then alternated.

The sites were periodically examined for barnacle settlement, as well as germlings of the alga, *Fucus gardneri*. The number of barnacle juveniles and germlings were recorded during each inspection. Each 100 cm² area was also photographed.

During the 1991 field season, this experiment will be expanded to include three additional study site pairs. Each site will be measured and prepared in a manner identical to that described above. All sites will continue to be monitored, counted and photographed during 1991.

Grazing by Limpets:

Based upon results obtained in 1990, this study will be discontinued.

Algal Succession:

At the termination of the limpet grazing experiment, an opportunity was presented to gain an understanding of temporal algal succession with Herring Bay. Several questions regarding algal succession in general are relevant to possible impacts from oil. These questions include:

1. Can a basic algal succession for Herring Bay be defined?
2. Based upon the number of sites chosen for study, is there a difference in algal succession (and thus, species composition) between reference and impacted sites?
3. Is there a difference in algal succession between areas that are simply scraped free of all algae, vs. the application of a killing agent (such as bleach)?

Because eight study sites were chosen for the limpet study (four pairs of impacted and reference sites) and the termination of the experiment involved removal of the fences and all algae within each 625 cm area, this provided a platform to easily examine algal species succession at multiple plots.

At the termination of the limpet grazing study, fences from two elevation contours were removed. Only the marine epoxy used to hold the fences in place was left to serve as a marker for each of the algal succession study plots.

All algae was removed from each 625 cm area. A coin was flipped to determine if the odd or even numbered fences would receive application of sodium hypochlorite (bleach). The results from this random selection are listed as follows:

Site #	Contour:	Date conducted
1411C	upper: Odd	9-10-90
1411C	lower: Odd	9-10-90
1311X	upper: Even	9-10-90
1312C	upper: Even	9-10-90
1312X	upper: Odd	9-11-90
3811C	upper: Even	9-12-90
3611X	upper: Odd	9-13-90
1713C	upper: Even	9-14-90
1713X	upper: Even	9-14-90
1251X	Odd	9-15-90
1251C	Even	9-16-90
1231X	Even	9-15-90
1231C	Even	9-17-90
1852C	Odd	9-18-90

Sessile invertebrates, such as barnacles were not removed from the 625 cm area. A photograph of each experimental was taken (with photolabels in each frame).

Each study plot will be revisited to assess algal species composition. At the time of visitation, percent cover of each plot by separate species will be determined by a point grid method. Also, photographs of each plot will also be taken, and a voucher specimen of each plant observed within each plot will be collected. Voucher specimens will be sent to UAF for taxonomic identification. Finally, numbers and species of invertebrate grazers found within the 625 cm area will also be counted.

This process will be repeated until four time series plots have been established for both the scraped and bleached plots at each contour. These study plots will be monitored quarterly.

Temperature and Salinity:

Stations adjacent to all 1990 study sites were monitored weekly for temperature and salinity at the 0, 0.5 and 1 through 5 m depth contours around MHHW, using a CTD meter. This procedure will be continued through the 1991 season.

APWG
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Study Title: Technical Support Study for the Restoration of
Dolly Varden and Cutthroat Trout Populations in
Prince William Sound

Study ID Number:

Lead Agency: Alaska Department of Fish and Game

Cost: \$147K

Inclusive Dates: March 1, 1991 - February 28, 1992

Project Leader Date Andrew G. Hoffmann
333 Raspberry Rd, Anchorage AK 99518
(907) 267-2195

Organization Date Chuck Meacham
Leader 333 Raspberry Rd, Anchorage AK 99518
(907) 267-2112

Financial Officer Date 333 Raspberry Rd, Anchorage AK 99518

INTRODUCTION

Information collected by NRDA programs during oil years 1 and 2 has documented injury to Dolly Varden and cutthroat trout in Prince William Sound (PWS). Mortality rates of Dolly Varden and cutthroat trout from oiled sites were significantly lower than control sites and there was also a highly significant reduction in the growth of cutthroat trout from oiled sites. Dolly Varden and cutthroat trout are both important to the recreational fisheries in PWS and these fisheries offer a diverse and often unique range of angling opportunities.

There were limited data available on the stock status of Dolly Varden and cutthroat trout prior to the oil spill although there are harvest and effort data available for the sport fishery through the Statewide Harvest Survey (SWHS) (Mills 1989). We also know through tag recoveries in 1990 that there is fishing mortality attributed to the by catch of Dolly Varden and cutthroat trout in the commercial fishery for salmon although the full extent of the by catch is not known. The additional mortality from oil perturbation has caused concern that some of the oil impacted stocks may be unable to sustain historical levels of fishing mortality. Reduction in growth due to oil perturbation may result in additional mortality and/or changes in size composition that impact historic fishing patterns on these stocks.

Due to these stock conservation concerns, the Department may have to enact restrictions on the sport fishery in the oil impacted areas which would greatly reduce the opportunities available to anglers. The most effective method to enhance the recovery of the oil impacted stocks is to redirect angling effort to populations outside of the oil impacted areas. This also provides alternative fishing opportunities for anglers who were displaced due to management actions taken to ameliorate impacts of the oil spill. At the present time there is a paucity of data available on the stock status of Dolly Varden and cutthroat trout populations outside of the oil impacted areas. It is imperative to collect the necessary data on these populations before we can redirect angling effort. It is also important not to exasperate the stock status of Dolly Varden and cutthroat in PWS by redirecting effort on populations of fish that cannot sustain increased sport harvest.

There is need to develop a consistent and rational approach in developing fisheries outside of the oil impacted areas. The sites of these fisheries will be selected according to a process that addresses a number of criteria. Each candidate water will be examined against a list of criteria to determine its suitability for promoting fishing opportunities.

At the conclusion of this project, a number of fishery locations will be identified outside of the oil impacted areas that will

provide the sport fishing public with a range of desired angling opportunities. The range of angling opportunities will be categorized into three zones based on remoteness and accessibility. This information will be disseminated to the angling public through brochures and other activities by the proposed Public Information restoration project. Additionally, data collected from this project could be used to identify further restoration opportunities for these stocks such as identifying possible sites for the placement of fish passes or critical habitat areas that could be protected through the purchase of private inholdings or mineral rights.

OBJECTIVES

The overall goal of this project is to develop a strategy to direct Dolly Varden and cutthroat trout sport fishing effort away from the oil impacted areas and into non-oiled locations in Prince William Sound. This goal will be met by accomplishing the following objectives and associated tasks.

Objective 1:

Identify stream systems in non-oiled locations in PWS that support viable populations of Dolly Varden and cutthroat trout.

Task:

- A. Inventory known locations of Dolly Varden and cutthroat trout in the non-oiled locations in PWS and categorize into three sport fishing zones based on remoteness and access.
- B. Visit a subset of sites identified and sample for Dolly Varden and cutthroat trout during the peak of the sport fishery (July through September) to determine fish catchability and attain a subjective estimate of stock size.
- C. Prepare a matrix of fishery characteristics to evaluate each stream system for potential as an alternative sport fishery for Dolly Varden and cutthroat trout fisheries outside the oil impacted area of PWS.

Objective 2:

Evaluate stock structure of overwintering populations of Dolly Varden and cutthroat trout at two sites in each of the three sport fishing zones. (Eyak and Culross lakes are examples of likely candidates in 1991).

Task:

- D. Estimate abundance of Dolly Varden and cutthroat trout in two sites within each of the three sport fisheries zones. (tests will be done at a precision level of $0.1 \pm 50\%$).
- E. Estimate the length composition of Dolly Varden and cutthroat trout in two sites within each of the three sport fisheries zones. (± 10 mm of their true value 90% of the time).

Objective 3:

Based on results from objectives one and two, make recommendations for locations to promote redirection of sport fishing effort outside of the oiled areas of PWS.

METHODS

This study consists of a five year plan to evaluate a variety of Dolly Varden and cutthroat trout systems in non-oiled locations in PWS to determine their potential for development as a sport fishery. In order to accomplish this a screening and evaluation process was developed (Figure 1). The first step in this process is the inventory of stream systems that support populations of target species. The characteristics of these systems will be evaluated relative to sport fisheries development using a matrix (Figure 2) to compare and categorize systems. Next, stocks of Dolly Varden and cutthroat trout will be assessed in certain streams selected using the matrix. Finally a management strategy will be developed for redirecting sport fishing activity away from the oil impacted area. This information will be made available to the public through a separate restoration project "Public Information" proposed by the Department of Fish and Game.

IDENTIFICATION OF DOLLY VARDEN/CUTTHROAT TROUT SYSTEMS

An inventory of potential Dolly Varden and cutthroat trout systems will be developed using available information and field reconnaissance. Based on remoteness and access each stream will be placed into one of three zones representing the type of sport fishing experience expected. Zone one would consist of systems accessible by road. Zone two would be those areas accessed by short boat trips limited to day trips. Zone three would be systems accessible only by extended overnight boat trips or by float plane.

Information from departmental and other agency studies, and other outside sources will be used to identify sites currently known to support Dolly Varden. Systems currently being used by sport fishermen will be identified using data from the Sport Fish Division's Statewide Harvest Surveys which provides harvest and effort data on specific sport fisheries in PWS. Other data sources include: lake surveys done in PWS by FRED division for PWS Aquaculture Corporation in the early 80s, inventories of anadromous streams by Habitat Division, and stream rehabilitation work done by the US Forest Service, and US Fish and Wildlife Service. These data will be reviewed to compile information on historical locations of Dolly Varden and cutthroat trout in PWS. Interviews will also be conducted with others knowledgeable of the fisheries in PWS, such as fishing guides, fishermen, and hatchery personnel to collect anecdotal information about potential systems. Field reconnaissance will be made at selected sites to determine the presence of target species and conduct an initial evaluation of stocks.

The field reconnaissance will involve visiting stream systems once or twice during the periods of peak sport fishing (July through September). At each site hook and line sampling will be conducted to determine the presence and catchability of stocks present. In addition, a subjective estimate of stock size and length

STRATEGY TO SELECT STREAMS TO REDIRECT SPORT FISHERY OUT OF THE OIL IMPACTED AREA

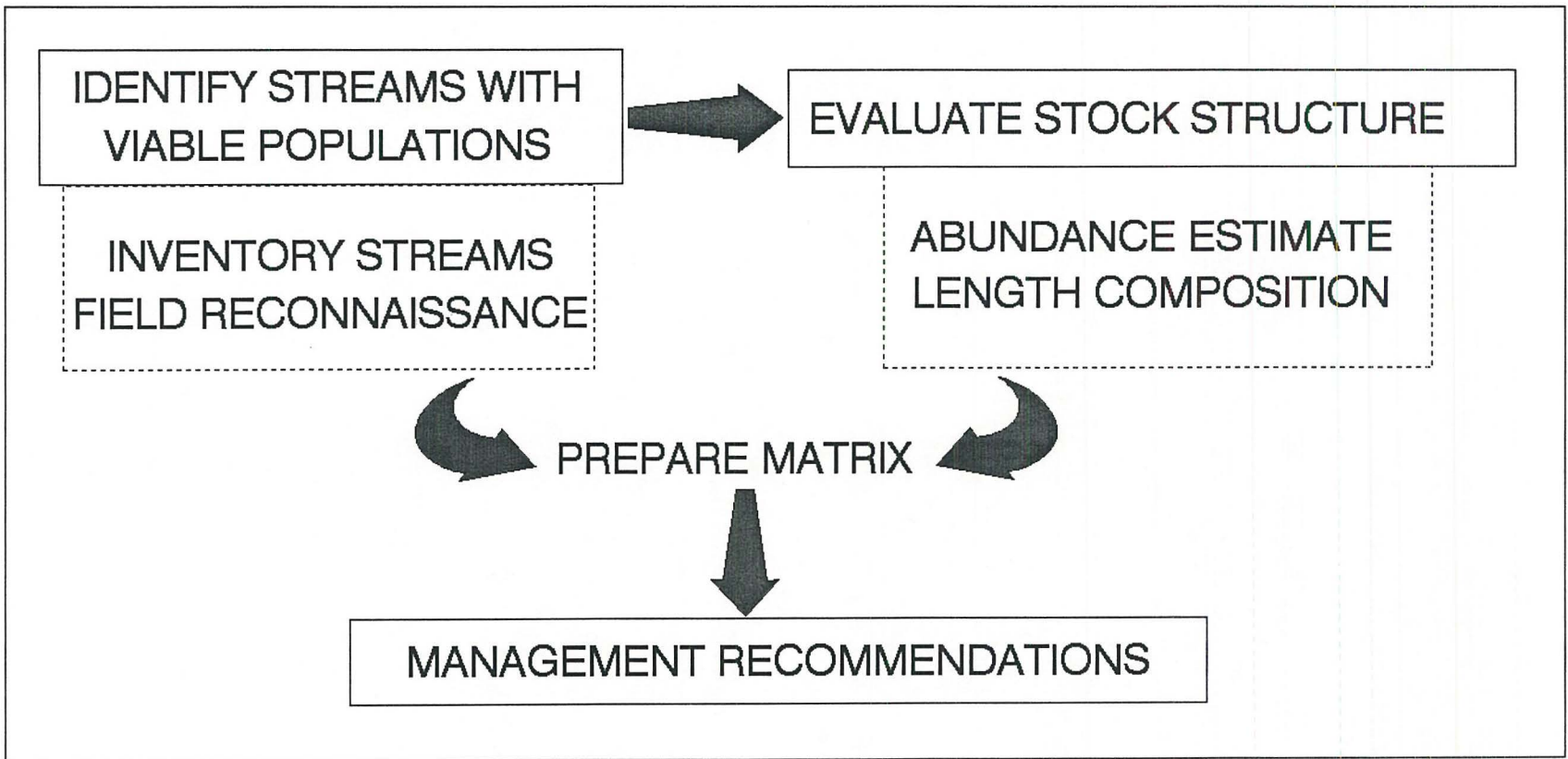


Figure 1. Strategy to select streams to redirect sport fishery out of the oil impacted area.

SCHEMATIC DIAGRAM OF MATRIX OF FISHERY CHARACTERISTICS

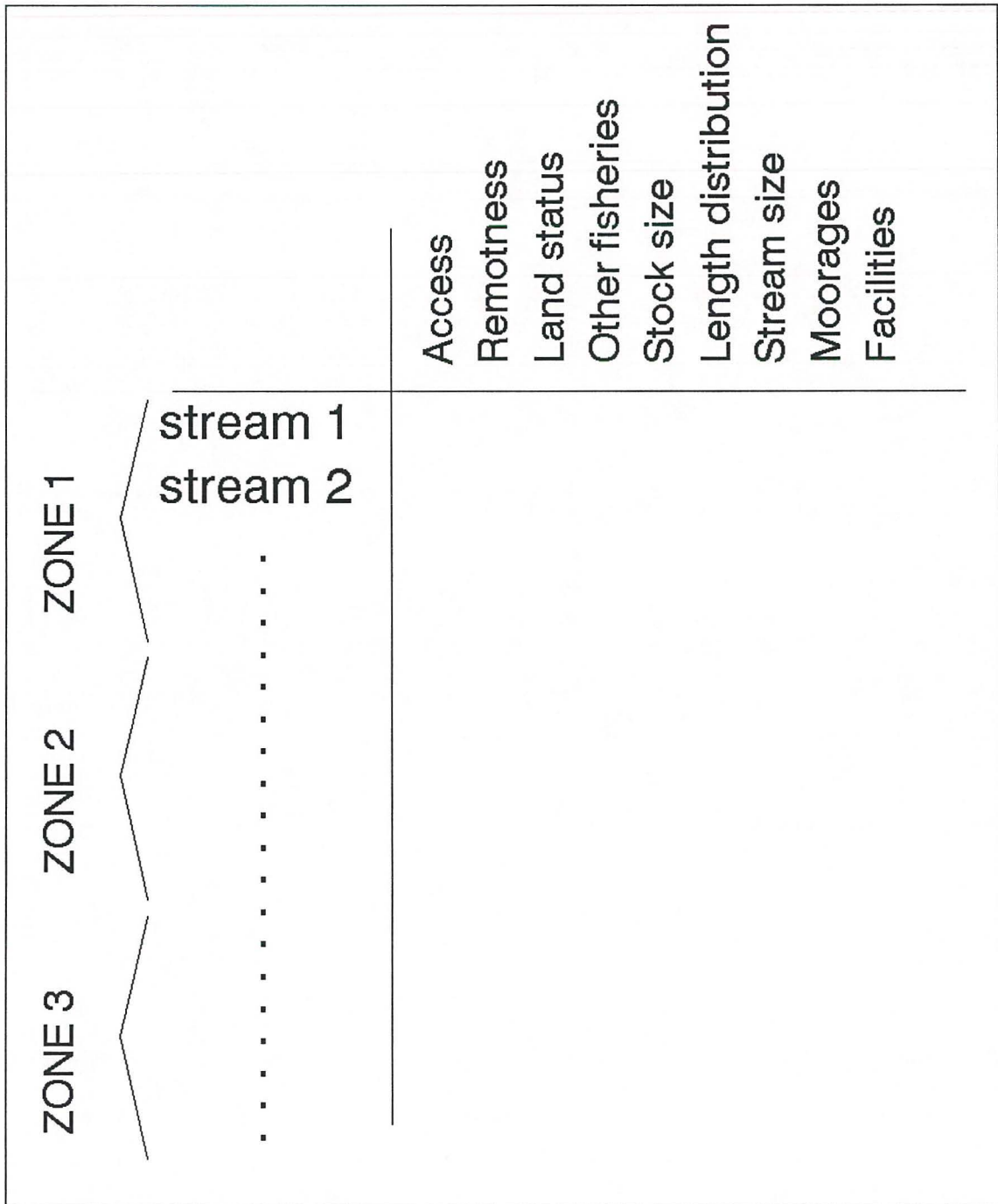


Figure 2. Schematic diagram of matrix of fishery characteristics.

distribution will be made by collecting fish using seines or electrofishing. All fish captured will be tagged with individually numbered tags. The purpose of the tagging is to allow for tracking of the movements of various populations to determine overwintering sites. The site will also be evaluated for the potential of conducting abundance estimates.

From the data collected in the inventory and reconnaissance a matrix of streams and associated characteristics will be developed to compare and categorize all streams identified as potential systems for Dolly Varden or cutthroat trout sport fisheries. Stream characteristics to be considered include access, remoteness, land status, proximity to other fisheries, stock size, length distribution of fish, unique characteristics of fishery, current and historic fishery, and physical nature of the stream.

STOCK ASSESSMENT

Estimates of abundance and mean length will be made at two suitable sites in each zone. Estimates of abundance will be made using mark recapture experiments. Weirs or fyke nets will be used as capture techniques depending on the nature of the stream system.

In systems selected for abundance estimates where the stream is suitable for constructing a weir an aluminum picket weir will be installed. Weirs will be placed approximately 0.5 km from the saltwater terminus of the streams upstream of tidal influence. During the spring sampling, weirs will be used to count and sample the emigration of Dolly Varden and trout from study streams. The weirs will be operated by a two-person crew from mid-April to early July. Downstream live traps will be installed.

In systems selected for abundance estimates where a lake is present and the stream is not suitable for a weir, fyke nets will be used. Lightweight, 3'X 3'X 13' fyke nets will be fished around the perimeter of the lake and around the outlet stream. During the spring sampling, fyke nets will be used to count and sample the emigration of Dolly Varden and trout from study streams. The fyke nets will be operated by a three person crew from mid-April to early July.

Each fish captured using either capture technique will be identified, counted, and measured (tip-of-snout to fork-of-tail to the nearest mm) and tagged using individually numbered Floy tags. Adipose fins will be clipped on all tagged fish as a secondary mark. Fish will also be examined for missing adipose fins, tags, or tag scars from other tagging studies. Scale smears will be collected from the preferred area from all cutthroat trout and placed individually on acetate slides in coin envelopes. Date, species, sex (if identifiable from external maturation characteristics), and length will be recorded for each fish.

During the second year of study and each subsequent year, each fish

containing a tag, a tag scar, or missing its adipose fin will be considered one recapture event. Recapture events will be recorded separately for fish containing tags and fish with missing tags. Recaptured fish with missing tags will be retagged. Fish with no visible tag scar and containing their adipose fin (not tagged in first year of study) will also be tagged. Tag numbers will be recorded for each recapture and each fish tagged.

All fish mortalities will be examined for presence of tags and adipose fins, identified, and measured as outlined above. Sex and maturity will be determined by internal examination, and sagittal otoliths will be collected. Date, species, sex, length, maturity, and tag number will be recorded. Fish containing tags, tag scars or missing adipose fins will be recorded as recaptures.

DATA ANALYSIS

Estimates of abundance will be computed for each study site through analysis of tag returns. The Jolly-Seber three-sample method (Seber 1982) will be used to estimate abundance at each study site. Buckland's program RECAP (1980) will be used to generate the estimates and variances. Abundance estimates will be used to approximate potential yields thus determining the level of sport fishing effort each system can support.

Length distribution data will be evaluated using Relative Stock Densities (RSD) as described by Gabelhouse (1984). Proportions of each species within specific length categories will be calculated for each system.

Recaptured data, in addition to being used for the abundance estimates, will be used to track the movements of fish. Tagging and recapture locations will be compared to determine movements among various systems and to determine overwintering areas.

SCHEDULES AND PLANNING

Year 1	Identification and inventory of Dolly varden and cutthroat trout systems.
Year 2-4	Reconnaissance and stock assessment field work.
Year 5	Development of management strategies.

BUDGET

Salaries	\$	80.0
Travel		2.0
Contracts		20.0
Supplies		30.0

Equipment	<u>15.0</u>
Total	\$ 147.0

PERSONAL QUALIFICATIONS

Andrew Hoffmann holds a bachelors degree in biology and a masters degree in environmental science/aquatic biology. He has worked with the Department of Fish and Game for ten years on a variety of projects. The work most relative to this project is his involvement as the assistant to the principle investigator for the NRDA study involved with the injury of the oil spill to the Dolly Varden and cutthroat trout in Prince William Sound. This experience has allowed him to become familiar with the Dolly Varden and cutthroat trout in the sound as well as gaining first hand experience with the field work, data analysis and administration of the project upon which this proposal is based.

RPWG
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PROPOSED OIL SPILL RESTORATION STUDY

I. Cover

Title: Identification of Upland Habitats Used by Marbled Murrelets in Prince William Sound

Study Identification Number: To be Assigned

Name of Study Leaders: Kathy Kuletz

Lead Agency: U.S. Fish & Wildlife Service
Marine and Coastal Bird Project
Anchorage, Alaska

Cooperating Agency: U.S. Forest Service
Glacier Ranger District
Girdwood, Alaska

Cost of Proposal: \$124,200

Inclusive Dates of Study Plan: March 1, 1991 - February 28, 1992

Signatures

Study Leader: _____

Supervisor: _____

Oil Spill Coordinator: _____

Financial Officer: _____

II. Introduction

The marbled murrelet (Brachyramphus marmoratus), a small nearshore alcid, is a species of concern from Alaska to California. They were listed as threatened in British Columbia in 1990 and are being considered for threatened or endangered status throughout its range in the United States. Loss of nesting habitat is postulated as the reason for their decline in B.C., Washington, Oregon and California. Population estimates for murrelets are not available for

all of Alaska, but the area affected by the oil spill is believed to be a population center in Alaska (Mendenhall 1988). Marbled murrelets suffered direct mortality from the Exxon Valdez oil spill disproportionate to their numbers at risk (Piatt et al. 1990). Preliminary analysis suggest a non-significant decline in summer populations in Prince William Sound (PWS), compared to pre-oil spill estimates. The winter population declined significantly along oiled shorelines compared to unoiled shorelines (Laing pers. comm.). In 1989, the Naked Island area showed population declines compared to pre-spill numbers, possibly due to human disturbance causing temporary displacement (Kuletz 1990a). Murrelets collected in the spill zone in August 1989, were exposed to petroleum hydrocarbon contaminants (Robinson-Wilson, pers. comm.), suggesting the possibility of long-term effects.

Recovery of the murrelet population could be enhanced by ensuring the availability of undisturbed nesting habitat. The Restoration Planning Work Group, building on expert and public input, identified protection of upland nesting habitats as one way to assist the natural recovery of species which depend on upland habitats for some stage of their life cycle. To fulfill this objective, specific information is needed on habitat requirements of the marbled murrelet.

Unlike most other seabirds, there are no conspicuous sites used by large numbers of nesting murrelets. Murrelets are secretive and widely-scattered (non-colonial) during their breeding season. In lower latitudes, the birds nest in coastal old-growth conifers (Marshall 1988, Nelson 1990, Quinland and Hughes 1990). In Southcentral Alaska nesting requirements are unknown. There are qualitative accounts of tree nesting but no nests have actually been found. However, several ground nests have been found, some of which could have been the closely related Kittlitz's murrelet (B. breverostris).

In 1990, a restoration feasibility pilot study investigated methods of studying upland use by marbled murrelets on Naked Island. Using information obtained in 1990, this proposal presents a study plan to assist in the identification of murrelet nesting habitat and specific areas of nesting activity in PWS.

III. Objectives

- A. Refine the censusing protocol for marbled murrelets at upland sites in Prince William Sound.
- B. Document tree nesting of marbled murrelets in Prince William Sound.
- C. Determine the presence and absence of marbled murrelet in selected upland habitat sites in Prince William Sound.
- D. Describe habitat associations in documented use areas in Prince William Sound.

IV. Methods

Objective A: Refining censusing protocol for murrelets in Alaska

Through all aspects of this study, information will be collected that will help establish guidelines for conducting upland habitat surveys of nesting murrelets. The influence of weather, seasonal patterns and observational techniques will be considered. The 1990 feasibility project at Naked Island (Kuletz 1990b) occurred from 9 June to 18 August. The standard survey is done by field personnel conducting a "dawn-watch survey." To determine variability in detections and seasonal patterns, three of the stations which were surveyed at least three times in 1989 will be surveyed at least bi-monthly in 1991. In 1990, three types of the "dawn-watch" survey developed by the Pacific Seabird Group (Paton et al. 1989) were attempted. The "intensive" dawn-watch survey (observer remains in one location) proved most suitable for the remote, uneven terrain of Naked Island and will be the basic field method of determining upland murrelet activity in 1991.

A dawn-watch survey is done at a pre-selected site during peak murrelet activity, when birds fly to their nests to exchange incubation duties or feed chicks. Since Naked Island birds displayed the same pattern as those at lower latitudes, each survey will be 45 minutes before to 75 minutes after official sunrise. Weather and lighting conditions (using a photography light meter) will be noted. Observers will use a tape recorder to note time of observation, type of detection (audio, visual or both), number of birds, number and types of vocalizations, direction and distance from the observed, and murrelet behavior (flight patterns, height of bird). Because birds may pass over an area without nesting there, certain behavioral activities and height of the bird will be used to classify the station as a "documented use area" (Nelson 1990). Birds flying silently through or circling below tree canopy, landing in trees or making stationary calls from trees indicate a documented use area.

All observers will be trained prior to the surveys, particularly in the classification of "detections" and in identification of murrelet calls and flight patterns. Field personnel will receive training via videos and audio tapes in the Regional Office, and in the field with the Study Leader. Training could begin as early as April, using sites in Kachemak Bay. Training at the Naked Island sites will not begin in early or mid-May.

A pilot study will be implemented to test the efficacy of self-activated tape records in determining murrelet activity in upland areas. If operable under Alaskan conditions, this system would enable greater coverage of areas where the number of field personnel are limited and access is difficult. The tape recorder will be set to record during the period of a dawn-watch survey in conjunction with a field observer. Test surveys will be made with the recorder at different heights, in clearings and in the trees. Data from field observers and the tape recorder will be compared for similarities in the number of audio visual detections.

Objective B: Documentation of tree nests in southcentral Alaska

In 1990, several sites with high murrelet activity were located on Naked Island. In some cases, potential nest sites were narrowed down to a few trees and birds were observed to land in trees. Now that these areas are known, a major effort to locate nests will be made on Naked Island using the proven "intensive ground survey" method (Naslund 1990, Singer et al. in press). Multiple observers (2-3), connected by hand-held radios, will focus on specific clusters of trees, and eventually individual trees, to locate a potential nesting branch. Once a suspected nest branch is located, a tree climber will climb an adjoining tree to document the nest. Data will be taken on nests following the Pacific Seabird Groups Nest Site Sampling Protocol (Varoujean and Carter 1989).

The search for nests will be augmented by use of audio equipment which can detect the soft calls made at the nest by adults and juveniles, and the wing beats of birds landing in trees (Singer, pers. comm., Nelson, pers. comm.). A portable cassette tape recorder, equipped with headphones and parabolic reflector will be used at documented use sites or suspected nest areas.

Objective C: Determine presence and absence in selected sites

The relationship between at-sea counts of murrelets and their upland nesting areas is unknown. This study will test for a correlation between at-sea densities and upland activity on a coarse scale. Results will indicate if at-sea counts are a reliable indicator of nearby upland nesting by murrelets. If so, future efforts to locate "documented use areas" can be more readily focused.

The presence and absence of marbled murrelets will be determined using intensive dawn-watch surveys. Based on results of boat surveys of waterbirds in PWS (Laing, unpubl. data), two types of shoreline sections, those with high murrelet densities and those with low murrelet densities, will be selected as focal points for dawn-survey stations. A survey station will be established approximately 200 meters inland from the middle of the shoreline transect. Data from Naked Island in 1990 indicated that numbers and flight direction relative to major habitat features could be determined. Among the seven paired stations in 1990 (each with a site near shoreline and another further inland), the amount of murrelet activity near the water was correlated with activity further inland (Kuletz 1990).

A minimum of 10 inland survey sites will be selected in both high and low density areas. Each site will be visited three times between May and mid-August, with surveys separated by at least two weeks at a given site (Nelson, pers. comm.). Field personnel will conduct dawn-watch surveys as described in Objective A, such that pertinent data on local habitat-murrelet associations will be available.

These surveys will require a mobile crew of at least two people, supported by a 25-foot vessel for transport around the PWS. Allowing for weather days and logistics, one crew could complete the 60 surveys required for the minimum number of survey sites. Increasing the sample size will require a second crew of two observers, which could be supported by the same boat.

Objective D: Describe habitat associations in documented use areas

Because Naked Island has known use sites, efforts to describe plant associations on a fine scale will be conducted by the U.S. Forest Service (USFS). At documented nesting sites already mapped, a plant-association crew will conduct ground surveys to provide detailed habitat data. The USFS has received \$40,000 to conduct this aspect of the study. This effort could be expanded with a second two-person dawn-watch survey crew which would be assigned pre-selected survey sites throughout Naked Island. The survey sites would be selected randomly within four habitat types defined by analysis of aerial photographs. The USFS will also provide maps of timber types occurring on Naked Island and other sites in the PWS.

V. Data Analysis

A. Tests

Data collected during 1991 will be combined with data collected in 1989 and 1990 and analyzed using standard statistical protocols.

B. Products.

This study will provide maps, computerized data sets and a final report on marbled murrelet activity at all surveyed sites. Detailed data on habitat and timber types will be compiled for all documented use sites and nest sites, through the cooperation of the USFS. The presence and absence of murrelets will be correlated with habitat. These data can be used in subsequent phases of the study to test predictions of murrelet presence in the field.

VI. Schedules and Planning

A. Report Submission Schedule:

March-April 1991	Prepare for field season/hire personnel
May-August 1991	Conduct field work

Sept.-Nov. 1991	Data input and analysis
December 1991	Draft report completed
February 1992	Final report completed

B. Sample and Data Archival.

Original copies of field data will be archived in the USFWS oil spill file system. Copies of the data set will be archived with the USFWS marine and Coastal Bird Project and the USFS Glacier Ranger District.

C. Management Plan

Kathy Kuletz will serve as the Study Leader or principal investigator. Ms. Kuletz works under the direct supervision of the Project Leader, Marine and Coastal Bird Project, Division of Migratory Bird Management, Fish and Wildlife Service, Anchorage, Alaska. The Study Leader is responsible for coordinating the completion of field data collection (including the habitat association information, analysis of field data; and timely reporting of the information in draft and final reports. The Project Leader is responsible for achieving coordination with all other marine bird studies during the planning, implementing, and reporting phases of the study. The USFS investigators are responsible for completing the habitat association descriptions and timber typing as described in this proposal. The USFS investigators work under the general direction of the USFWS Study Leader, all of whom will cooperate toward the accomplishment of the study objectives.

D. Logistics

To complete this study will require the use of a 25-foot vessel and field camps in Cabin Bay on Naked Island and other appropriate locations in the PWS.

VII. Budget

Salaries and Overtime

Study Leader GS-11 (1FTE)	-	\$ 48,000
Biotech GS-7 (1FTE)	-	35,000
Biotech GS-5 (.4FTE)	-	9,100
Biotech GS-5 (.4FTE)	-	<u>9,100</u>
Total Salaries		\$101,200

Travel/Per Diem	-	\$ 7,000
Supplies	-	6,000
Equipment	-	<u>10,000</u>
Total		\$124,200

IX. CITATIONS

- Kuletz, K.J. 1990a. Assessment of Injury to Waterbirds from the Exxon Valdez Oil Spill: Effects on Populations of Marbled Murrelets Along the Kenai Peninsula and Prince William Sound. Draft report, bird Study 6. U.S. Fish and Wildlife Service, Anchorage, Alaska.
- Kuletz, K.J. 1990b. Restoration Feasibility Study No. 4: Identification of Upland Habitats Used by Wildlife Affected by the EGOS: Marbled Murrelets. Draft report. U.S. Fish and Wildlife Service.
- Marshall, D.B. 1988. Status of the Marbled Murrelet in North America with special emphasis on populations in Washington, Oregon and California. USFWS Biol. Rep. 88(30).
- Naslund, N.L., S.W. Singer and S. Singer. 1990. A proposed ground search technique for finding tree nests of the marbled murrelet in open canopy forests. Abstract, Pacific Seabird Group Symposium, Feb. 22-25, 1990, British Columbia.
- Paton, P.P.W., C.J. Ralph, H.R. Carter and S.K. Nelson. 1989. The Pacific Seabird Group's 1989 handbook for Marbled Murrelet surveys at inland sites. Unpubl. report, USDA Forest Service, Pacific Southwest Research Station, Arcata, CA.
- Piatt, J.F., C.J. Lensink, W. Butler, M. Kendziorek, D. Nysewander. 1990. Immediate Impact of the 'Exxon Valdez' Oil Spill on Marine Birds. *Auk* 107(2):387.
- Quinlan, S.E. and J.H. Hughes. 1990. Location and Description of a Marbled Murrelet Tree Nest Site in Alaska. *condor* 92:1068-1073.
- Singer, S.W., S. Singer and N. Naslund. 1991 (in press). Discovery and observations of two tree nests of the Marbled Murrelet. *Auk*.
- Thomas, D.W. 1988. The distribution of bats in different ages of Douglas-Fir forests. *J. Wildl. Manage.* 52(4):619-626.
- Varoujean, D.H. and H.R. Carter. 1989. The Pacific Seabird Group's Marbled Murrelet nest site sampling protocol. Main and Estuarine Research company, 2269 Boardway, North Bend, OR.

STATE OF ALASKA

DEPARTMENT OF FISH AND GAME

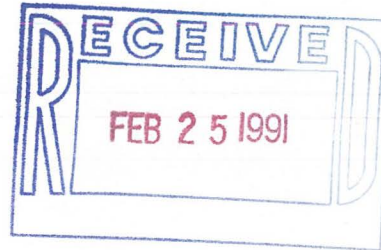
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WALTER J. HICKEL, GOVERNOR

1300 COLLEGE ROAD
FAIRBANKS, ALASKA 99701-1599

RPW/G
N

February 21, 1991

Roy Nowlin
OSIAR Division
Alaska Department of Fish and Game
Anchorage, AK



Dear Roy:

Enclosed is a restoration science study proposal for harbor seals entitled "Habitat Use, Behavior, and Monitoring of Harbor Seals in Prince William Sound." The proposal covers the period March 1, 1991 - February 28, 1993 or State Fiscal Years 1991, 1992 and 1993. As requested, budgets are broken down by oil spill period as follows: March 1-June 30, 1991; July 1, 1991-February 28, 1992; March 1-June 30, 1992; and July 1, 1992-February 28, 1993. The proposal combines the two brief restoration proposals submitted last winter into one integrated package that includes satellite tagging to study behavior and haulout use and aerial surveys to monitor recovery from declines caused by the EVOS. Since aerial surveys will be flown in 1991 as part of my NRDA study (we hope), this proposal includes only one set of surveys in September 1992. If additional surveys to monitor recovery are considered desirable, they will be proposed at a later date.

While the cost of this work may appear relatively expensive, it represents a substantial cost savings due to cooperation and collaboration with the National Marine Mammal Laboratory (NMML), the Alaska Sea Grant Program, and Texas A&M University. Software for PTT microprocessors has already been developed for NMML sea lion transmitters and we will also be able to use data analysis software developed for sea lions. NMML has donated the cost of data acquisition for the first five transmitters to be attached in 1991, as well as provided us with satellite ID codes. Randy Davis's salary is being paid by Texas A&M. Sea Grant is allowing Kate Wynne to work on this project at no additional cost, and she in turn is enlisting the volunteer help of Cordova fishermen.

The proposal has been prepared in Word Perfect 5.1. A diskette is enclosed. If you have any questions, please call. I plan to be in the office all of next week.

Sincerely,

Kathy Frost
Marine Mammals Biologist
Division of Wildlife Conservation

cc: D. Calkins
W. Regelin
T. Loughlin
S. Sanner ✓

I. COVER PAGE

**Exxon Valdez Oil Spill Restoration Science Study
Detailed Study Plan**

Title: Habitat Use, Behavior, and Monitoring of Harbor Seals in Prince William Sound

Study ID Number:

Project Leader: Kathryn J. Frost

Lead Agency: Alaska Department of Fish and Game

Cooperating Agency: NOAA/National Marine Fisheries Service

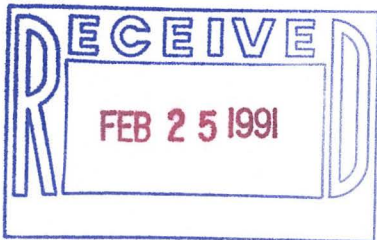
<u>Cost of Proposal:</u>	SFY 91 (March-June 1991)	\$ 46,500
	SFY 92 (July 1991-June 1992)	\$164,400
	SFY 93 (July 1992-February 1993)	\$151,700

Dates of Study: 1 March 1991-28 February 1993

Project Leader: Kathryn J. Frost 2/21/91
 Kathryn J. Frost
 Alaska Department of Fish and Game
 1300 College Road
 Fairbanks, AK 99701
 phone (907) 456-5156

Organization Leader: _____

Financial Officer: _____



21 February 1991

II. INTRODUCTION

The semi-enclosed waters of Prince William Sound (PWS) provide very good habitat for harbor seals (Phoca vitulina) and other marine mammals. Several thousand seals occur in PWS, where they are commonly seen hauled out on rocks, reefs, beaches, and glacial ice. Harbor seals are used for subsistence by residents of coastal communities such as Tatitlek, Chenega, and Cordova. Tourists and recreational users of PWS enjoy watching and photographing harbor seals. They, like other marine mammals, are protected by provisions of the Marine Mammal Protection Act. Because of a population decline that is going on in PWS and other parts of Alaska, it is possible that other protective legislation such as the Endangered Species Act may be invoked to provide for conservation and recovery of harbor seals.

Harbor seals were impacted by the Exxon Valdez oil spill (EVOS) in PWS. They encountered oil in the water and on haulouts. Early in the spill several fetuses, pups, and older animals were found dead in the impacted area. Studies conducted as part of the Natural Resources Damage Assessment (NRDA) program documented a substantial decline in the number of seals in oiled areas.

The number of seals in PWS had been declining prior to the EVOS. Twenty-five haulout sites in eastern and central PWS have been used to monitor trends in abundance since 1984. The mean number of seals in the trend count area during late summer surveys declined by 40% between 1984 and 1988, from 1,796 seals to 1,058 seals, a rate of about 10% per year (Pitcher 1989). Subsequent to the spill, the population decline continued at about the same rate at unoiled locations. However, at oiled sites the decline was much greater. From 1988 to 1990 harbor seals in the oiled portion of the trend count area declined 35%, compared to 13% in unoiled areas (Frost 1990).

Because of the decline in harbor seals, which was exacerbated in the area impacted by the EVOS, it is particularly important to understand what factors are limiting the population. We cannot assume, given the ongoing decline, that the number of seals in oiled areas will return naturally to pre-spill levels. It is necessary both to continue monitoring population trends and to identify and appropriately manage areas of particular biological significance in order to augment recovery in any way possible. Most of the information currently available on harbor seals in PWS consists of counts of animals on haulouts during pupping and molting. While these data are essential for monitoring changes in overall abundance, they are not adequate for determining what is causing the seal population to decline, or for designing conservation and management measures to facilitate recovery and ensure its future health. There is no information available on site fidelity, movements between haulout sites, seasonal changes in hauling out patterns, habitats used for feeding, or feeding behavior.

Recently developed satellite-linked telemetry can be used to gather information on all of these important aspects of harbor seal biology. Miniature platform transmitter terminals (PTTs) have created opportunities to monitor location and diving behavior of marine mammals (Mate 1986; Hill et al. 1987; Mate 1989; Stewart et al. 1989; R. Merrick personal communication). The PTTs transmit to a satellite-based Doppler positioning system that calculates locations and tracks movements of animals with considerable accuracy. When combined with appropriate environmental sensors and microprocessor hardware and software, other information about an animal's environment and behavior can be transmitted to the satellite.

The goals of this study are to gather data on the behavior and habitat use of harbor seals in PWS that can be used to design effective conservation measures, and to monitor the abundance and trends of harbor seals at trend count sites in oiled and unoiled areas of PWS using standardized methodology. Habitat use and behavior studies will be conducted by attaching satellite transmitters to harbor seals at selected sites, and determining their movements, diving patterns, feeding locations, and haulout patterns. Population monitoring will be conducted by flying aerial surveys of the trend count route during the autumn molt. Counts will be compared to data collected prior to and during the EVOS in order to document whether and how rapidly natural recovery occurs. In 1991, monitoring will be included as part of NRDA Marine Mammal Study No. 5, Assessment of Injury to Harbor Seals in Prince William Sound, Alaska and Adjacent Areas. In 1992 and any future years, monitoring will be included as part of this restoration study.

III. OBJECTIVES

1. To describe haulout behavior of satellite-tagged harbor seals in PWS relative to date, time of day, and tide.
2. To describe the use of particular haulouts by satellite-tagged harbor seals in PWS and the frequency of movements between haulouts.
3. To describe patterns of movements of harbor seals within PWS and between PWS and adjacent areas.
4. To describe diving characteristics and feeding behavior of harbor seals in different habitats in PWS.
5. To use the data provided by this study to identify important harbor seal habitat and recommend management actions necessary to safeguard that habitat.
6. To use data provided by this study to interpret aerial survey data and to refine aerial survey methodology.

7. To monitor harbor seal population trends at trend count sites in oiled and unoiled areas of PWS and to determine whether seal numbers in areas impacted by the EVOS recover to pre-spill levels.

IV. METHODS

A. Habitat use and behavior

We propose to begin the investigation of harbor seal habitat use in PWS with a pilot study in which five satellite-linked PTTs will be attached to seals at either Applegate Rocks or Bay of Isles. Two seals will be caught and instrumented during April 1991 and three others in September 1991. Information obtained from these tagged seals will be used to evaluate tag performance and to determine baseline values for parameters such as depth of dive and dive duration. This information will allow future tags to be programmed with appropriate default values and threshold levels such that they will gather and store the maximum amount of useful data.

In April-May 1992, after preliminary data have been evaluated, 10 harbor seals will be tagged at Applegate Rocks, Bay of Isles, and Olsen Bay in Port Gravina. These areas differ in regard to habitat type and degree of oiling during the EVOS. An additional 10 seals will be tagged in September 1992, after the molt, again at Applegate Rocks, Bay of Isles, and Olsen Bay. Seals will be caught by entanglement in nets placed near the haulouts. Transmitters will be attached to the back of the seal by gluing with epoxy resin (Fedak et al. 1984). The transmitters should remain attached for several months or until the following autumn when they will be shed during the molt.

Data will be acquired from the ARGOS satellite receiving system and analyzed using software provided by the manufacturer of the transmitters. Each PTT (approximate size 15cm x 15cm x 3cm) will transmit geographical locational information to the to a polar-orbiting satellite whenever an uplink to the satellite occurs. This will happen when the seal is hauled out or when it surfaces sufficiently long for transmission to occur and the satellite is positioned to receive the signal. Units will also be equipped with built-in programmable microprocessors to collect and summarize data for periods when animals are diving and store it for later transmission, as has been done for crabeater seals (Hill et al. 1987) and Steller sea lions (R. Merrick, personal communication). These data will be stored in memory until the seals haul out on land. A sea water switch will indicate when the animal is hauled out and all data stored in memory will then be transmitted during the next satellite overpass. Dive data will be summarized as histograms and dive profiles. Temperature sensor data will also be reported.

Each PTT broadcasts a unique identification code so that data can

be assigned to a particular seal. Position accuracy for all geographical locational information is rated by Service ARGOS to reflect the predicted accuracy of the calculated locations (Fancy et al. 1988, Stewart et al. 1989). Data acquired for harbor seals in this study will be screened for accuracy and interpretation of results will take into account signal quality. Sensor data will be used to validate whether the animal was at sea or hauled out on land when data were acquired, since errors in calculated locations may falsely indicate that a seal is on land or at sea (see Stewart et al. 1989).

Data on the haulout patterns of tagged seals will be examined for indications of daily or seasonal variations, for example to determine whether there is a change in the frequency of haulout by season, or whether the amount of time spent hauled out changes. Plots of locations where continuous signals are received will be used to determine the degree and regularity of use of particular haulout sites. We expect to receive fewer locations of seals while at sea, because the transmitter's antenna will frequently be submerged. However, at-sea locations will be plotted as an indication of areas used for feeding. Information on depth and pattern of diving will be compiled, and can provide some additional information on the general areas used for feeding.

These data will be used to evaluate site fidelity of seals, to quantify the amount of interchange among haulouts within and outside of the area impacted by the EVOS, to determine seasonal importance of particular haulouts, and to identify areas used for feeding. This information will help to identify areas of particular biological significance, and will serve as the basis for management recommendations to ensure the integrity of important seal habitats. They will also be valuable in further refining aerial survey methodology, particularly in determining the best time to conduct surveys.

B. Monitoring

In September 1991 we plan to conduct aerial surveys of harbor seals in PWS as part of the third and final year of NRDA Marine Mammal Study No. 5. Future monitoring of the status and trend of the harbor seal population in PWS should be conducted as part of this restoration science study to determine whether harbor seals have recovered from declines caused by the EVOS. Surveys will follow a trend count route previously established by ADF&G (Calkins and Pitcher 1984; Pitcher 1986, 1989). The trend count route covers 25 haulout sites and includes six sites that were impacted by the EVOS (Agnes, Little Smith, Big Smith, Seal, and Green islands, and Applegate Rocks), 16 unoiled sites, and three intermediate sites that were not physically oiled but were adjacent to oiled areas (Table 1). Visual counts will be made of seals at each site and photographs taken of large groups for later verification. Seven to 10 replicate daily flights will be made, timed such that counts are

within two hours of low tide. The survey protocol will be the same one used in 1989 and 1990, as shown in Appendix I.

V. DATA ANALYSIS

A. Behavior and Habitat Use

Locations calculated by Service ARGOS will be screened for accuracy and plotted on charts of PWS to preliminarily classify whether the seal was on land or a sea. Locational data will be compared with sensor data, when possible, to verify that these classifications are correct. Patterns of diving and hauling out will be presented as histograms. Dive data histograms will present the number of dives at different depth increments and by duration of dive. Means and standard deviations for dive depth and duration will be calculated and compared for seals in different locations or habitats and at different times of year.

Dive data will be presented as graphs and histograms which indicate the range in individual behavior as well as summary data for all seals combined. Dive profiles will be plotted graphically and examined to identify dive patterns, for example deep feeding dives or shallow dives indicative of travelling. Compilation of data on time and location of feeding dives will be used to identify feeding areas near different haulouts. Dive and haul out cycles will be examined relative to time of day, tide, and season. Haulout bouts and tidal cycles will be overlaid and plotted. Summaries of the number and quality of uplink data and at-sea position data will be presented in tabular form. Tabular summaries will also be prepared for use of different haulouts by individual seals; the number of haulout bouts relative to tidal state and time of day; and frequency of haulout and amount of time spent feeding by season.

B. Monitoring

Harbor seal surveys must be conducted within biological time windows imposed by the molting period. Sample size for aerial surveys is partly determined by weather which can limit flight altitudes. While results of previous harbor seal trend counts have indicated that it is desirable to obtain 7-10 counts during a survey period (Pitcher 1986, 1989), in actuality the number of counts is almost always limited by the number of days suitable for flying.

Aerial surveys do not estimate the total number of seals present since they do not account for seals that are in the water or seals hauled out at locations not on the trend count route. Surveys provide indices of abundance based on the number of hauled out seals counted. Interpretation of trend count surveys relies on the assumption that counts of harbor seals on select haulout sites are valid linear indices of local abundance. We assume that within a given biological window, such as the molting period, haul out

behavior remains the same from one year to the next, and counts can thus be compared. Standardization of procedures minimizes the affects of variables such as tide and weather that could influence the number of seals hauled out on a given day. Behavioral data obtained from satellite transmitters attached to seals as part of this study will help to verify these assumptions.

Reliable surveys of the trend count route were conducted during the molt in 1984, 1988-1990, and will also be done in 1991. These data will be used for comparions with data collected after 1991. Analysis of monitoring data and comparisons with other years will be conducted following statistical methodology used for previous molting surveys (Frost 1990).

Overall trends in abundance during the autumn molt, and trends at oiled versus unoiled sites, will be examined using a repeated measures ANOVA (Winer 1971) performed on the trimean (Hoaglin et al. 1985) of the site count data for September surveys. The trimean statistic will be used as a measure of central tendency because sets of counts at a single location sometimes show bimodal distributions or include extreme variations. The test assumes that the mean proportion of the population hauled out on the trend count route is constant over years. Orthogonal contrasts derived from the ANOVA will be used to compare average counts in oiled and unoiled areas (see Frost 1990 for detailed description of contrasts and analyses).

VI. SCHEDULES AND PLANNING

A schedule of field activities, data analysis, and report preparation is presented in Table 2 and a list of key personnel in Table 3. Field trips to attach PTTs will take place in April and September of 1991 and 1992. Trend count surveys during the molt will be conducted during September 1991 as part of an NRDA study and in September 1992 as part of this study. Field progress reports will be submitted within 30 days of the completion of each field effort. These reports will be in letter form and will summarize dates and activities during the field effort; personnel involved; location and number of seals tagged; the status of signal monitoring; and a brief summary of findings. Data retrieval and analysis will be ongoing throughout the period when PTTs are transmitting data. An interim report will be submitted by December 31, 1991 which will describe progress to date and present the preliminary results in the form of charts, histograms, graphs, and tables. The final report will be submitted by February 28, 1993. It is the intent of the investigators to prepare the results of this study for publication in the peer-reviewed literature after completion of the project.

Satellite data and survey data will be archived at ADF&G in digital format. Hard copy will also be generated and filed at ADF&G and a copy sent to the National Marine Mammal Laboratory. Copies of

digital satellite data will also be held Texas A&M University. All data will be organized and filed according to standard scientific procedures. Original copies of field data will be retained at ADF&G and copies provided to the Trustees upon request. Copies of study plans, data analyses, summaries, and reports will also be filed at ADF&G.

The project will be coordinated and managed by ADF&G. Cooperators will included Texas A&M University, the Alaska Sea Grant Marine Advisory Program, the National Marine Mammal Laboratory (NMML), and Cordova District Fishermen United. Application of satellite tags in 1991 will be done under authority of NMFS permit number 584, issued to the NMML. ADF&G will request a permit to authorize application of 20 tags in 1992.

Identification codes and data aquisition for the five PTTs applied in 1991 will be provided by the NMML. In 1992, ADF&G will be responsible for procuring identification codes and retrieving data. Data analyses will be conducted by personnel from ADF&G, with cooperation and assistance from Texas A&M University.

The Alaska Sea Grant Marine Advisory Program and members of Cordova District Fishermen United will assist by providing some logistics and support of field activities.

Software for programming the PTT microprocessors and for data analysis will be provided by the tag manufacturer. If necessary, additional data analysis software will be developed or acquired by ADF&G.

VII. BUDGET

A line item breakdown of costs from March 1991 through February 1993 is as follows:

<u>Line Item</u>	<u>Cost</u>			
	<u>Mar-Jun 91</u>	<u>Jul-Feb 92</u>	<u>Mar-Jun 92</u>	<u>Jul-Feb 93</u>
100 Personnel	10,200	43,000	13,200	61,100
200 Travel	3,700	5,000	4,000	7,000
300 Services	7,100	32,000	6,700	39,300
400 Commodities	25,500	45,000	5,500	44,300
500 Equipment	0	10,000	0	0
TOTAL	46,500	135,000	29,400	151,700

A detailed breakdown of these costs is presented in Appendix II.

VIII. PERSONNEL QUALIFICATIONS

Kathryn Frost has conducted research on marine mammals in Alaska since 1975. She has undertaken research on natural history and ecology of seals and beluga whales, including aerial and

photographic surveys of seals and whales; radiotagging of beluga whales to study behavior and movements; and studies of food habits and trophic interactions of seals, belugas, walruses, and bowheads. She has conducted extensive aerial surveys of harbor seals in PWS and boat-based observations and sampling of harbor seals as part of damage assessment studies following the EVOS. She is currently conducting a study of the habitat use and haulout behavior of spotted seals in northwestern Alaska, and is initiating a program to attach satellite tags to spotted seals.

Lloyd Lowry is the Marine Mammals Coordinator for the State of Alaska. He has conducted research on marine mammals in Alaska since 1975, including studies of the natural history, ecology, distribution, abundance, and food habits of seals, walruses, and whales. He participated in the EVOS response, and damage assessment studies on harbor seals. He participated in the development and application of radiotags for beluga whales. He has been responsible for project coordination and management of state and federally funded research projects, and is familiar with the federal marine mammal permit system.

Kate Wynne has conducted research on harbor seals and other marine mammals since 1981. She has worked in PWS since 1988 and is familiar with the area and its marine mammal populations. She has worked closely with area residents, particularly fishermen, since 1989 in documenting marine mammal-fishery interactions and in developing awareness of marine mammal issues and concerns. She has had previous experience catching and attaching radiotags to harbor seals and testing prototype satellite tags for walruses. She has conducted harbor seal, sea otter, and whale surveys in PWS and other parts of Alaska.

Randy Davis has conducted research on the biology and physiology of marine mammals since 1976. He specializes in the diving behavior and physiological adaptations for diving in marine mammals and penguins. His research has included field and laboratory studies of swimming energetics, including the swimming metabolism of harbor seals; under-ice movements of antarctic seals; and the effects of oil on sea otters. He has used radio telemetry and time depth recorders in his studies.

IX. CITATIONS

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X. OTHER INFORMATION

Appendix I. Standard methodology for harbor seal trend counts.

The most generally used methodology for enumerating pinnipeds is by aerial surveys of hauled out animals. The objective is to conduct the surveys at a time when a relatively large and consistent proportion of the population is hauled out and can be counted. Pinniped haulout patterns may be affected by a large number of

factors, including weather, time of day, tidal stage, and disturbances.

In the case of harbor seals, maximum numbers haul out during the pupping period (May-June) and during the molt (August-September) (Pitcher and Calkins 1979; Calambokidis et al. 1987). Availability of most haulout sites is limited by tidal stage, therefore more animals are usually hauled out a lower stages of the tide. Survey flights are therefore timed to coincide with daylight low tides, starting within two hours before low water and finishing within two hours after low tide.

In order to provide statistically valid estimates of the average number of seals hauled out in a trend count area, a number of sites are counted repetitively. The sites are selected so that they form a route that can be flown within a four hour period. The trend count route in Prince William Sound includes 25 haulout sites. Statistical considerations indicate that it would be desirable to obtain 7-10 replicate counts at each site during the survey period. In practice the number of counts may be limited by the number of days with good weather during the survey period (which is limited by the seasonal behavior patterns of the seals).

Surveys are usually conducted from a single engine fixed-wing aircraft (e.g., Cessna 180 or 185). Haulout sites are flown over at an altitude of 200-300 meters and seals are photographed with a hand held 35-mm motor driven camera with a 70 to 210-mm zoom lens. High speed (ASA 400) film is used. Color slides are commercially developed and the seals are counted from images projected on a marlite screen. Visual counts or estimates of seal numbers are also made while haulouts are being circled.

Data are tabulated by individual haulout site. If there is a reason to suspect that a particular count is not valid (e.g., haulout empty with a boat nearby) it is not included in the analysis.

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EVOS Harbor Seal Restoration Study, 1991-1993

Appendix II. Detailed budget breakdown for marine mammals restoration study "Habitat Use, Behavior, and Monitoring of Harbor Seals in Prince William Sound." Dollar amounts are in thousands.

		<u>Budgeting Period</u>								
		<u>Mar 91- June 91</u>		<u>Jul 91- Feb 92</u>		<u>Mar 92- Jun 92</u>		<u>Jul 92- Feb 93</u>		
<u>Line Item 100 - Personnel</u>										
<u>Person</u>	<u>Grade</u>	<u>\$/mo.</u>	<u>(mos)</u>	<u>\$</u>	<u>(mos)</u>	<u>\$</u>	<u>(mos)</u>	<u>\$</u>	<u>(mos)</u>	<u>\$</u>
K. Frost	WBIII	5.8	(1.0)	5.8	(3.0)	17.4	(1.0)	5.8	(4.5)	26.1
L. Lowry	WBIII	6.4			(0.5)	3.2	(0.5)	3.2	(1.0)	6.4
R. Delong	APIII	4.4	(1.0)	4.4	(2.0)	8.8			(1.5)	6.6
Clerical	CTIII	3.2			(1.0)	3.2			(1.0)	3.2
Technician	WTIV	4.2			(1.0)	4.2	(1.0)	4.2	(3.0)	12.6
D. Calkins	WBIV	6.2			(1.0)	6.2			(1.0)	6.2
TOTAL LINE ITEM 100				10.2		43.0		13.2		61.1
<u>Line Item 200 - Travel and Per Diem</u>										
Field Travel (Fbks or Anch to Cordova; Houston to Cordova)				2.6		2.9		2.9		3.4
Meeting Travel (Fairbanks to Anchorage)						0.7				0.7
Field Per Diem				1.1		1.1		1.1		2.6
Meeting Per Diem						0.3				0.3
TOTAL FOR LINE ITEM 200				3.7		5.0		4.0		7.0
<u>Line Item 300 - Services and Contracts</u>										
Data Aquisition (\$2,200/PTT)						22.0				22.0
Vessel Charter				6.0		6.0		6.0		6.0
Aircraft Charter										8.0
Printing, Copying, Graphics				0.5		0.8		0.2		1.0
Telephone				0.3		1.0		0.2		0.8
Air Freight and Postage				0.3		1.2		0.3		1.0
Equipment Repair						1.0				0.5
TOTAL LINE ITEM 300				7.1		32.0		6.7		39.3

EVOS Harbor Seal Restoration Study, 1991-1993

Appendix I. Continued.

	Budgeting Period			
	Mar 91- <u>June 91</u>	Jul 91- <u>Feb 92</u>	Mar 92- <u>Jun 92</u>	Jul 92- <u>Feb 93</u>
<u>Line Item 400 - Commodities</u>				
Satellite Transmitters	17.5	35.0		35.0
Seal Nets	2.5	2.5		1.0
Vessel Fuel	2.5	2.5	2.5	2.5
Film and Processing				0.5
Computer Supplies/Software	1.0	2.5	0.5	2.7
Field Supplies	<u>2.0</u>	<u>2.5</u>	<u>2.5</u>	<u>2.6</u>
TOTAL LINE ITEM 400	25.5	45.0	5.5	44.3
<u>Line Item 500 - Equipment</u>				
Outboard Motor		4.0		
Satellite Uplink Receiver		5.3		
Waterproof housing	—	<u>0.7</u>	—	—
TOTAL FOR LINE ITEM 500	0	10.0	0	0
GRAND TOTAL	46.5	135.0	29.4	151.7

Table 1. Prince William Sound harbor seal trend count route.

Site #	Description	Status relative to EVOS
1	Sheep Bay	unoiled
2	Gravina Island	unoiled
3	Gravina Rocks	unoiled
4	Olson Bay	unoiled
5	Porcupine Point	unoiled
6	Fairmont Island	unoiled
7	Payday	unoiled
8	Olsen Island	unoiled
9	Point Pellew	unoiled
10	Little Axel Lind Island	unoiled
11	Storey Island	intermediate
12	Agnes Island	oiled
13	Little Smith Island	oiled
14	Big Smith Island	oiled
15	Seal Island	oiled
16	Applegate Rocks	oiled
17	Green Island	oiled
18	Channel Island	intermediate
19	Little Green Island	intermediate
20	Port Chalmers	unoiled
21	Stockdale Harbor	unoiled
22	Montague Point	unoiled
23	Rocky Bay	unoiled
24	Schooner Point	unoiled
25	Canoe Passage	unoiled

EVOS Harbor Seal Restoration Study, 1991-1993

Table 2. Schedule of activities from April 1991 through February 1993 for restoration study "Habitat Use, Behavior, and Monitoring of Harbor Seals in Prince William Sound." Letters are initials of personnel indicated in Table 3.

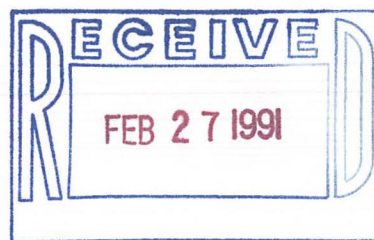
Activity	Dates	Personnel
Coordination meeting	11-15 March 1991	KF, LL, KW, RD
Procure five PTTs	March 1991	KF, RD
Reserve 1991 ARGOS satellite channels	March 1991	KF, RD
Program and test PTTs	March 1991	KF, RD
Attach two PTTs	10-25 April 1991	KF, LL, KW, RD
Submit field progress report	May 1991	KF
Data retrieval and preliminary analysis of satellite data	May 1991-February 1993	KF, LL, RD, RAD
Apply for NMFS tagging permit	May 1991	LL, KF
Program and test PTTs	August 1991	KF, RD
Attach three PTTs	10-30 Sept. 1991	KF, LL, KW, RD
Submit field progress report	October 1991	KF
Procure ten PTTs	November 1991	KF
Prepare interim report	November-December 1991	KF, LL, RD
Submit interim report	31 December 1991	KF
Reserve 1992 ARGOS satellite channels	January 1992	KF, DR
Program and test PTTs	March 1992	KF, RD
Attach ten PTTs	April 1992	KF, LL, KW, RD
Submit field progress report	May 1992	KF
Procure ten PTTs	May 1992	KF
Conduct trend count aerial surveys	August-September 1992	DM, KF
Program and test PTTs	August 1992	KF, RD
Attach ten PTTs	September 1992	KF, LL, KW, RD
Submit field progress report	October 1992	KF
Analyze aerial survey data	October-December 1992	KF, EB, RAD
Final data analysis	December-February 1993	KF, LL, RD, EB, RAD
Prepare final report	January-February 1993	KF, LL, RD
Submit final report	28 February 1993	KF

Table 3. Personnel involved in restoration study "Habitat Use, Behavior, and Monitoring of Harbor Seals in Prince William Sound."

Name	Affiliation	Responsibilities
Kathryn Frost	ADF&G	Project leader; tagging; aerial surveys; data analysis; reporting
Lloyd Lowry	ADF&G	Project review and coordination; assist with tagging, data analysis, and reporting
Earl Becker	ADF&G	Consult on biometrical procedures
Don Calkins	ADF&G	Project review and coordination
Randy Davis	Texas A&M	Assist with tagging, data analysis, and reporting
Robert A. DeLong	ADF&G	Data analysis
Tom Loughlin	NMFS/NMML	Project review; data acquisition
Dennis McAllister	ADF&G	Assist with aerial surveys
Dan Reed	ADF&G	Satellite data acquisition
Kate Wynne	UA Sea Grant	Field coordination; assist with tagging

RPWG
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I. COVER PAGE



A. Title: Prince William Sound Harlequin Duck
Breeding Habitat Analysis

B. Study ID Number (assigned by RPWG)

C. Project Leader: Dr. Samuel M. Patten

D. Lead Agency: Alaska Department of
Fish and Game

Cooperating Agency: U.S. Fish and
Wildlife Service

E. Cost of Proposal: 225K

F. Inclusive Dates of
Study Plan:

Mar. 1, 1991
to
Feb. 28, 1992

G. Signatures: Name: Date: Telephone:

Project Leader:

Organization Leader:

Organization Financial Officer:

II. INTRODUCTION

This is a study plan, proposed by the Division of Wildlife Conservation of the Alaska Department of Fish and Game, for restoration of harlequin duck (Histrionicus histrionicus) populations in Prince William Sound (PWS) as a result of the Exxon Valdez Oil Spill (EVOS) of March 24, 1989. Harlequin ducks are both resident in, and winter migrants to Prince William Sound (Isleib and Kessel, 1973). They feed in intertidal zones which were heavily impacted by the EVOS, and breed along nearby streams (Hogan, 1980).

Preliminary estimates from boat surveys conducted in 1989 by the U.S. Fish and Wildlife Service indicate a summer population of approximately 6000 harlequin ducks in Prince William Sound. This number is substantially augmented by winter migrants from northern and interior montaine breeding areas. An estimated 10,000 harlequin ducks winter in Prince William Sound (Isleib and Kessel, 1973).

Harlequin ducks, because of their resident status and intertidal foraging habits, have been considered substantially at risk to effects of the EVOS (King and Sanger, 1979). Harlequin ducks have recently (1990) been listed as an endangered species in eastern Canada. Reasons given for this listing include over-hunting and chronically low productivity (Goudie, pers. comm.).

Harlequin ducks are dependent upon intertidal marine invertebrates (Vermeer and Bourne, 1982). Harlequins consume a wide variety of small mussels, clams, snails, chitons, limpets, salmon and herring eggs (Koehle, Rothe and Dirksen, 1982; Dzinbal and Jarvis, 1982). Bivalves, particularly blue mussels (Mytilus), and small clams (Macoma), are noted for their ability to concentrate pollutants at high levels (Shaw et al, 1976). The crude oil spilled from the Exxon Valdez has caused severe damage to marine invertebrates that support harlequin ducks (Stekoll, Clement, and Shaw, 1980) and bioaccumulation in the food chain is resulting in uptake of petroleum hydrocarbons by harlequin ducks over a long period (Dzinbal and Jarvis, 1982; Sanger and Jones, 1982).

Harlequin ducks were subject to considerable initial direct mortality resulting from the EVOS. NRDA Bird Study No. 11 has also documented levels of petroleum hydrocarbon ingestion by sea ducks, including PWS Harlequins, with resulting physiological and life-history effects (Hall and Coon, 1988; Patten, 1990). In addition to direct mortality associated with the EVOS, Patten (1990) showed a significant proportion of the Harlequin population surviving in oiled areas of PWS is in physiologically poor condition. This is associated with consumption of oiled intertidal prey items. Affected birds exhibit minimal adipose tissue and concentrations of petroleum chemicals and metabolites in liver and bile. Results of

the summer 1990 investigation of resident harlequin ducks in western Prince William Sound further indicate a reproductive failure and population decline in the oil spill area. In contrast, a stable population and normal reproduction was observed in summer 1990 in unoiled areas of Prince William Sound.

Little is known about Harlequin Duck breeding parameters in Prince William Sound other than they nest along forested streams. Several studies have been conducted elsewhere on the breeding ecology of the Harlequin Duck, one in Iceland (Bengston, 1966) and another study in Glacier National Park, Montana (Kuchel, 1977). Habitat utilization by Harlequin Ducks has been studied in Grand Teton National Park (Wallen, 1987). The Idaho Department of Fish and Game has recently conducted a series of studies of Harlequin Duck breeding distribution and abundance along streams in that State (Wallen and Groves, 1988, 1989, Cassirer 1989, Cassirer and Groves, 1989, Cassirer and Groves, 1990).

Specific information is lacking about Harlequin Duck breeding in Alaska. Dzinbal's (1982) MS thesis on ecology of Harlequin Ducks in Prince William Sound during summer and Dzinbal and Jarvis' (1982) work on summer coastal feeding ecology provided limited data on specifics of Harlequin breeding biology in Alaska.

USFWS and ADF&G biologists attending the initial EVOS Restoration Planning meeting in Anchorage (April 3-4, 1990) identified the lack of knowledge of Harlequin Duck breeding habitat ecology in Prince William Sound as being a critical data gap which needs to be addressed as part of restoration efforts for this species. Increase in knowledge about Harlequin breeding ecology received a priority rating at that meeting. Restoration of Harlequin Duck populations in oiled areas of Prince William Sound will likely depend upon productivity of resident Harlequin Ducks remaining in unoiled sections of Prince William Sound. Harlequin nesting streams in Prince William Sound will need special protection from impending logging, aquaculture, mariculture, and hydroelectric projects if this seaduck population is to recover from the results of the 1989 Exxon Valdez Oil Spill.

As a result of public and agency suggestions received at a series of EVOS restoration planning meetings, a Harlequin Duck Restoration Feasibility Study was conducted by ADF&G in summer 1990 in Prince William Sound. The goals of this feasibility study were to locate Harlequin nesting streams, describe breeding habitats, and where possible obtain initial breeding productivity indices. Information gathered during the 1990 field season demonstrates the 1991 Harlequin Duck restoration project is technically and logistically feasible.

Harlequin Ducks historically bred throughout Prince William Sound, including areas effected by the EVOS. No Harlequin broods were observed in the oil spill area in 1990. Harlequin breeding concentrations were noted in several areas of northeastern Prince William Sound; around Port Etches, Hinchinbrook Island; on southwestern Montague Island; and in northwestern Prince William Sound (College Fiord). The greatest concentration of nesting Harlequin ducks is apparently located in northeastern Prince William Sound, unfortunately in an area scheduled to be logged. Logging effects could significantly retard or thwart efforts to restore the population of Harlequin Ducks in Prince William Sound after the EVOS. Riparian forest zones or stream conservation easements should be protected as part of the "acquisition of equivalent resources" phase of the oil spill restoration program. Many wildlife species including harlequin ducks would benefit.

Concentrations of molting and flightless male harlequin ducks were noted inside the oil spill area of western Prince William Sound. Up to 70 individual ducks were observed in these concentrations. The largest of these aggregations was observed at Foul Bay, near the entrance to Port Nellie Juan. An characteristic of these molting sites was their location in extensive rocky intertidal zones in secluded bays. These sites appeared highly productive, were used by a variety of avian and mammal species, and may need protection as part of further restoration efforts.

III. OBJECTIVES

- A. To locate, identify and describe harlequin nesting streams in Prince William Sound.
- B. To identify habitats used by nesting harlequin ducks including stream, riparian, and adjacent forest types.
- C. To describe environmental parameters which may affect harlequin breeding, such as distance of the nest site from the coast, distance of the nest site from the stream, physical features of the nest site, and associated vegetative, limnological and hydrological characteristics of nesting streams. This will be done to address the following questions: what are the biological and physical features of the stream environment used by the female harlequin duck for nesting habitat selection? Why are some streams selected and others not?
- D. To measure harlequin breeding productivity, such as clutch size, hatching and fledging success, in oiled and unoiled areas of Prince William Sound.

- E. To identify, as management goals for this restoration project, a) which streams (i.e., those used by nesting harlequin ducks) in PWS require stream conservation easements in order to protect harlequin breeding sites from the effects of impending logging, and b) the size of forested buffer strips to delineate those easements.
- F. To identify potential alternative methods and strategies for restoration of lost use, populations, or habitat. Although harlequin ducks are believed to be ground nesters in Prince William Sound, we will investigate the concept that artificial structures such as nest boxes, affixed to trees, might raise harlequin productivity, as part of efforts to restore this population (Paige, pers. comm.).

IV. METHODS

The methods for this project are designed to answer the following questions: what are the biological and physical features of the stream environment used by the female harlequin duck for nesting habitat selection? Why are some streams selected and others not? These methods incorporate known technical feasibility and will not interfere with cleanup activities or ongoing NRDA studies.

Two field camps are planned for the 1991 season. The first camp is located in eastern Prince William Sound at Olsen Bay, Port Gravina, in an unoiled area. Private holdings in this area soon may be logged. The second camp is located on Knight Island in the oil spill area.

The anticipated harlequin duck restoration activities for 1991 include: 1) an extensive survey of anadromous streams and moulting sites around Prince William Sound; 2) an intensive study of known nest site parameters; and 3) the development of a predictive model of nest site selection, based on hydrological and limnological characteristics of their nesting streams.

1990 stream walks in oiled and unoiled areas of Prince William Sound will be replicated to locate and identify additional harlequin breeding habitats. The presence of harlequin pairs in spring at stream mouths suggests later breeding use of those streams. Nesting females feed at stream mouths and fly upstream to incubate clutches.

Harlequin females will be mist-netted and radio-tagged at selected stream mouths and later tracked along streams to nest sites in oiled and unoiled areas of PWS. Sample size goals for radio-tagging include 30 females from the oil spill area of western PWS and 30 females from the unoiled area of eastern PWS.

Nesting females are secretive and nests are otherwise difficult to locate. Although harlequin broods are regularly observed in Prince William Sound, no nests have yet been located. A 1.5g - 5g radio transmitter with a small, 3-month battery pack, epoxy-glued to the upper tail feathers of a nesting female, is recommended as the least intrusive method of radio-tagging these ducks. The outer layer of feathers will cover the transmitter; only the antenna will project. The tail feathers and the transmitter will be moulted from the female in late summer, after brood-rearing. A small radio transmitter, glued to the upper back feathers of a Marbled Murrelet, has successfully been used to track this salt-water diver to its forested nest site (Quinlan and Hughes, 1990).

Harlequin duck clutch size, hatching success, and brood size (a part of the productivity index) will be obtained from sample nest sites located by radio-tracking females. Hens with larger ducklings proceed from nesting streams to intertidal shorelines in late summer. Brood size (fledged ducklings/hen) can be readily counted.

Nesting habitats will be described qualitatively and quantitatively. This will include physical, chemical and biological characteristics of nesting streams as well as documentation of riparian and adjacent forest types. These measurements will facilitate development of a predictive model of Harlequin breeding habitat which will, in turn, suggest riparian areas for conservation easements, and answer the question as to why some streams are selected for nesting and others not.

Boat surveys of shorelines inside and outside the oil spill area will be conducted to locate intertidal moulting sites in July as well as census hens with broods in August. Post- or non-breeding males are concentrated while flightless during July in secluded bays with extensive rocky intertidal zones. Primary feather moult produces a period of maximum energy demands and is a vital stage in the life cycle. The flightless birds require substantial amounts of high-quality food, located in the immediate vicinity, for primary feather regrowth. Restoration of the harlequin duck population in Prince William Sound should include identification of important sites and some measure of protection for the areas. The intertidal moulting sites are believed highly productive, and are utilized by a variety of other species such as seals, sea otters, seabirds and shorebirds.

A. Sampling methods: Since nesting harlequin ducks feed on salmon eggs when available, these ducks are usually found along anadromous fish streams. Information concerning anadromous fish streams (where harlequins nest) is available from Commercial Fisheries Division of the Department of Fish and Game. Approximately 900 anadromous fish streams are located in Prince William Sound. An experienced observer from Commercial Fisheries Division reported Harlequin sightings while walking 140 of these streams in 1990. The streams were selected for investigation based upon three factors:

- 1) prior historical sampling for fish concentrations;
- 2) 80% of the PWS pink salmon production originates from these watercourses;
- 3) the streams are spatially distributed throughout Prince William Sound, including oiled and unoiled areas.

These 140 streams were walked during the summer of 1990, and information recorded on harlequin duck sightings and habitats. Additional 1991 data on Prince William Sound harlequin distribution will be requested from U.S. Fish and Wildlife Service biologists conducting aerial and boat surveys. Interviews will be conducted with Commercial Fisheries personnel working in Prince William Sound stream surveys in 1991. Other agency or private biologists working in Prince William Sound and having knowledge of harlequin ducks will be consulted in 1991. This data will complement 1991 information produced by boat and stream surveys associated with the Seaduck Damage Assessment Project (Bird Study No. 11) in the oil spill area of western Prince William Sound.

For discriminant analysis, a random sample of hydrological, limnological and vegetative data from PWS streams avoided by breeding harlequin ducks will also be required in order to compare with data from streams selected by nesting females.

B. Citations.

See IX. Citations section.

C. Standard Operating Procedure Requirements

See attached field S.O.P. for sampling
(Cassirer and Groves, 1990).

D. Quality Assurance and Control Plans:

Data will be recorded in standard formats.
Chain of custody procedures as outlined in
State/Federal Damage Assessment Plan Analytical
Chemistry QA/QC will be followed.

V. DATA ANALYSIS

A. Tests

The null hypothesis states that harlequin duck clutch size, hatching and fledging success in the oiled area of PWS are greater than or equal to those parameters in the unoiled area. The alternative hypothesis states that harlequin duck clutch size, hatching and fledging success are worse in the oiled area of PWS than in the unoiled area.

B. Analytical Methods

Discriminant analysis will be used to determine which variables characterize streams selected by nesting harlequin ducks (Srivastava and Carter, 1983). An exact Wilcoxon Rank Sum Test will be used to test differences in clutch size, hatching success, nest success, and brood size in oiled and unoiled areas of PWS: $F(x) \geq G(x)$; $F(x) < G(x)$ (Lehmann, 1975).

The proportion of successful Harlequin nests in the unoiled areas of PWS will be compared to those in the oiled area, using a one-sided t-test for proportions (D'Agostino et al., 1988).

C. Products

The products of this study will be a narrative report with maps, figures, and tables.

VI. SCHEDULES & PLANNING

A. Data and Report Submission Schedule:

Fieldwork	March 1, 1991 to Aug. 30, 1991
Analyze Data	Sept. 1, 1991 to Nov. 30, 1991
Complete Interim Report	Jan. 31, 1992
Complete Final Report	June 30, 1992

Special Reports:

Additional interim reports and communications will be prepared by the PI as a secondary priority.

B. Sample and Data Archival:

Samples and data will be archived at the Department of Fish and Game.

C. Management Plan

This study will be conducted and managed by the Principal Investigator who will work under the general guidance of a Division of Wildlife Conservation Oil Spill Management Coordinator. The Management Coordinator will provide general supervision during planning, implementation, and reporting phases of the study. The Principal Investigator and assistants will collect the field and laboratory data. The Principal Investigator will interpret results, and write draft and final reports. General guidance may also be provided by the DWC Waterfowl Coordinator. The Principal Investigator may be also assisted in the field by a number of DWC/UAF biologists, technicians, or graduate students.

D. Logistics

Field aspects of this study in Prince William Sound will be conducted from Whittier and Cordova, with the Department of Fish and Game facilities at Anchorage and Cordova providing a base of support. Operations will be conducted in oiled and unoled parts of PWS. A field camp was constructed at Herring Bay, Knight Island, in Summer 1990, in the oil spill area. Operations will resume at this site in Spring 1991. A field camp at Olsen Bay in eastern Prince William Sound will be constructed with U.S. Forest Service assistance in Spring 1991. This site will allow access to Harlequin nesting streams in the unoled area of eastern PWS. Access to each site will be by air charter or center console fiberglass boats. Three biologists/technicians will staff each site.

VII. Budget

A. Costs:

Salaries	\$125,000	(12 mo WBIII at 5600; 1ea
		3 mo WBII at 4150; 1ea
		1 mo WBIV at 6150; 1ea
		4 mo TIII at 2500; 4ea)
Travel	20,000	
Contracts	35,000	
Supplies	15,000	
Equipment	30,000	

Total	\$225,000	

Breakdown by Supplemental Appropriation (Until June 30, 1991).

Salaries	\$30,000
Travel	10,000
Contracts	10,000
Supplies	10,000
Equipment	15,000

Total	\$75,000

VIII. Personnel Qualifications:

1. Principal Investigator: - Samuel M. Patten

Sam Patten received his B.A. degree from Cornell University in 1968, majoring in Biology and German. He attended Heidelberg University 1968-71. In 1971 he began work as a Research Assistant at the University of Washington, conducting thesis research on Glaucous-winged Gulls in Glacier Bay National Monument under National Park Service sponsorship. He received his Master of Science degree in 1974.

He worked as a Research Associate for the University of Alaska in the summer of 1974, conducting research on avian populations on the outer coast of Glacier Bay for the National Park Service in an area potentially impacted by nickel mining. In 1975 he began research on gulls on the south coast of Alaska as a doctoral student at Johns Hopkins. Field work was conducted as part of the NOAA-OCS gas and oil baseline studies prior to the development of oil resources. He received his Ph.D. in Animal Ecology and Behavior from the Department of Pathobiology, School of Hygiene and Public Health, Johns Hopkins University, in 1980, with a dissertation on the evolution of gulls in Alaska.

Patten continued work on seabirds, shorebirds, and waterfowl in Yakutat, Alaska, for Operations Research, Inc., 1980-81, under NOAA contract. He assisted in production of a data atlas of the Bering, Chukchi, and Beaufort Seas for NOAA while at the University of Alaska 1981-82. He also conducted research on avian populations in the Susitna Basin, as part of the hydroelectric project, for the University of Alaska Museum in 1982. He began working for the Department of Fish and Game as Area Biologist for the Yukon-Kuskokwim Delta in 1983, conducting a cooperative management program instrumental in the population recovery of four species of geese. This management program also led to the expansion of muskox, moose, and caribou populations on the Yukon-Kuskokwim Delta through 1989. Since May 1989 he has been working as a Division of Wildlife Conservation researcher in the Oil Spill Impact Assessment and Restoration (OSIAR) program as a result of the Exxon Valdez Oil Spill.

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RPWG
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Proposed Oil Spill Restoration Study

I. COVER

Title: Population Status and Reproductive Success of Pigeon Guillemots and Black Oystercatchers in Prince William Sound

Study Identification Number: To be Assigned
Name of Study Leaders: Pigeon Guillemots - To be Assigned
Black Oystercatchers - Brad Andres

Lead Agency: U.S. Fish and Wildlife Service
Marine and Coastal Bird Project
Anchorage, Alaska

Cooperating Agency: None
Cost of Proposal: \$120,000
Inclusive Dates of Study Plan: March 1, 1991 - February 28, 1992

Signatures:
Study Headers: _____

Supervisor: _____
Oil Spill Coordinator: _____
Financial Officer: _____

II. INTRODUCTION

The pigeon guillemot (C^epphus columba) is a nearshore diving seabird which is highly vulnerable to oil spills (King and Sanger 1979). Guillemots were widely scattered in small colonies throughout the Exxon Valdez oil spill zone, and were subject to direct mortality from oiling (Piatt et al. 1990). ~~on several levels~~ ^{damage assessment} by different studies. Being a conspicuous and important member of the intertidal community in Prince William Sound, the black oystercatcher (Haematopus bachmani) was also studied during the oil spill. Black oystercatchers were documented to have been damaged by the oil spill. This proposal outlines a restoration feasibility study that will provide information on possible avenues of enhancing guillemot and oystercatcher reproductive success and develop a program to monitor recovery of each population.

Pigeon guillemots in Prince William Sound (PWS), which are at a 20 year low, declined significantly along oiled shorelines compared to unoiled shorelines (Laing 1990). There were declines of up to 52% along the Kenai Peninsula coast (Rice, pers comm.) and Kodiak as well (Laing 1990). Historic data was also available on a finer scale for the Naked Island area. Oakely (1989) and Kuletz (1990) surveyed guillemots at known colonies, and found the numbers significantly reduced along oiled shorelines compared to unoiled shorelines. Petroleum contaminants have been identified in adult tissues and unhatched eggs collected from Naked Island. Finally, in 1990, chick growth and fledging weight were the lowest of 6 recorded years (Kuletz 1990).

The guillemot's foraging and social behavior brings them into contact with shallow waters and intertidal rocks during the breeding season, exposing them to oil long after initial impact. Site-specific exposure at the colonies necessitates reliable data on colony location and size which can be compared among years.

Reproductive studies indicate that pigeon guillemots could experience long-term impact from the oil spill which could impair their natural recovery in the affected area. Information on guillemot diet and contaminate levels of their prey at different locations in the PWS could help determine the relative importance of prey type and oiling of forage areas to reproductive success.

At Naked Island, a primary source of nest failure was avian and mammalian predation. The extent of predation pressure in other areas of the PWS, and possible remediation of its impact, could assist in promoting recovery of the population. Currently, there is insufficient knowledge about the location and true size of major breeding sites, and whether the data on diet and reproductive success from Naked Island is representative of the PWS in general.

Reproductive success of black oystercatchers was studied on Green (an oiled site) and Montague islands (a non-oiled site) in 1989. Habitat uses at both sites were studied, and behavior of territorial pairs was studied on an opportunistic basis.

A preliminary analysis suggests that hatching success of black oystercatcher clutches was higher on Green Island, and that predators destroyed a larger number of nests on Montague Island. Chick survival appeared to be inversely correlated with the degree of oiling, with the lowest mortality on Montague Island (non-oiled site), higher mortality on moderately oiled territories on Green Island, and the highest mortality on heavily oiled territories on Green Island.

The average oystercatcher feeding rate (ingestion/minute) was approximately 2 1/2 times slower on Green Island than on Montague Island. Oystercatchers on Green Island did not spend a greater proportion of time preening, flying, or socializing than on Montague Island.

III. OBJECTIVES

A. Pigeon Guillemots

1. To identify the locations and determine sizes of pigeon guillemot colonies in the PWS.
2. To establish a censusing program for pigeon guillemots that will insure accurate and repeatable counts of guillemot colonies.
3. To determine reproductive success of pigeon guillemots and the causes of egg and chick mortality at Naked Island and other selected sites in the PWS.

B. Black Oystercatchers

1. To determine reproductive success of black oystercatchers and causes of egg and chick mortality at Green and Montague islands, Naked Island, and other selected sites in the PWS.
- 2/ To determine the density of black oystercatchers at Green, Montague, and Naked islands and other selected sites in PWS.

IV. METHODS

Objective A.1: Location and sizes of breeding colonies

Previous surveys of the PWS provide a base to identify general guillemot distribution, but because of the all-day censusing period, they miss some colonies and underestimate the size of others. Methods have been developed to census guillemots at their colonies during peak attendance with the least amount of variance (Nelson 1987, Ainley and Boekelheide 1990, Ewins 1990, Kuletz 1983). Peak attendance varies among geographic regions, but the patterns observed at Naked Island should be applicable to the rest of the PWS.

Guillemots will be censused by two observers per boat (inflatable or 25-foot whaler, depending on area) cruising selected shorelines from 50-100 meters offshore, in good weather, between 0430-0900 hours. Because guillemots at Naked Island extend their colony attendance during high tide, morning census times can be extended to 1000 hours if tides are appropriate. While this method will provide colony location and approximate colony size from early May to mid-August, variability can be reduced by censusing early in the breeding period (Ewins, pers. comm.). Preferably, early morning censusing will be conducted from early May to mid-June, to avoid missing incubating birds.

The survey areas will be distributed throughout the PWS relative to guillemot densities as indicated from previous surveys (Irons 1985, Laing 1990). Shoreline sections which had relatively high guillemot numbers, will be selected for censusing. During censusing, the number of guillemots, habitat and weather conditions will be recorded on field data sheets, using existing transect numbers for general reference. The exact location of colonies will also be assigned reference numbers and marked on maps.

Objective A.2: Establishing a censusing program for guillemots

This study will provide a database for specific guillemot colonies and areas of concentration, which can be used for a long-term monitoring program. In addition to the high density transects, a sample of shoreline transects which did not show guillemot concentrations in previous surveys will also be surveyed using methods described in Objective A.1. Results will determine if future guillemot censuses need to be more extensive, or if there is appropriate nesting habitat near large colonies will be identified and incorporated into the censusing effort (Ewins 1989).

Pigeon guillemots at Naked Island displayed colony attendance during the 2 hours around high tide, in addition to the early morning period (Kuletz 1983, unpubl. data). Where scheduling permits, colonies at other sites will be counted every half hour during daylight hours, from a good vantage point (preferably on land). If high-tide attendance is consistent throughout the PWS, future censusing efforts can utilize the hours near high tide.

Objective A.3: Locate nests and determine reproductive success

Guillemots nest in talus, rock crevices and cliff-top burrows. Because they are difficult to find, some aspects of reproductive success (i.e., nesting attempts and hatching success) depend on nests located in previous years. Nests are found by observing birds entering their burrows during the social periods prior to egg laying (early morning of high tide), or when the adults bring food to their chicks (primarily mid-July to mid-August).

Naked Island will be included in the 1992 study due to the advantage of having 4 pre-oil and 2 post-oil years of data for this site. Most importantly, there are marked nests at 11 colonies on the island for which nesting attempts and hatching success can be determined. This area will be visited at least once in late June, to record nesting attempts, and again in late July/early August to obtain fledging success and weights. Other sites in the PWS, known to have high concentrations of guillemots, will also be explored to locate nests.

During the pre-egg-laying censuses, field personnel will attempt to locate nests on Naked Island and other selected sites in the PWS, and will note which colonies have accessible nest sites. In July, field personnel will return to these colonies and continue nest searches by observing birds returning with food. Nests will be marked and mapped, and accessibility described and categorized. Previously marked nests on Naked Island will be checked for hatching success. In late July and early August, fully feathered chicks at Naked Island will

be weighed with pesola scales. At this time, any indication of predation can be obtained by egg and chick remains. Where possible, chick feces will be collected for identification of fish otoliths. It would also be possible at this time to collect unhatched eggs for contaminate analysis.

Objective B.1: Determine reproductive success

Nests will be located and marked on Green, Montague, and Naked islands and other selected sites in the PWS. Nests located on Green and Montague during 1989 will be relocated (Sharp 1990). Naked Island nests, previously found during other studies on the island will also be relocated. Other sites such as west Knight Island will also be explored for nests. Standard reproductive measurements will be collected (e.g., phenology, clutch size, chick growth rates, and hatching and fledging success.

Observations of adult behavior and foraging behavior will also be conducted and compared to data collected during 1989.

Objective B.2: Determine densities

Shoreline surveys of Green, Montague, and Naked islands will be conducted at biweekly intervals. Other sites in the PWS will be selected and surveys conducted but at less frequent intervals. Because the extent of various shoreline habitats differ at the sites (and oystercatcher density may differ in each type), shoreline habitats will be mapped. If still visually apparent, oil in the intertidal zone will also be recorded at systematic intervals during mapping as light (0-33%), moderate 34-66%) or heavy (67-100%). During a shoreline survey, the following information will be recorded: number, location, status (single, paired, paired with young, flock), evident color-marks. Average oystercatcher densities and variances (birds/km) will be estimated using simple random sampling formulae. Comparisons of bird density among oil categories will be made for each shoreline habitat. Differences in bird density will also be examined by status group (single, pair, pair with young). Because large numbers of shorebirds have been recorded on Montague Island (Norton et al. 1990), all other species of shorebirds encountered during surveys will be noted.

IV. DATA ANALYSIS

A. Tests.

Data collected during 1991 will be combined with data collected in 1989 and 1990 and analyzed using standard statistical protocols.

B. Products.

This project will provide tables and maps identifying guillemot and oystercatcher numbers in the study areas. Oystercatcher nest sites and guillemot colony sites will also be mapped. Sites suitable for future censusing and reproductive studies will be identified and nests marked on-site. Fish otoliths will be identified in the lab to provide a list of prey species used at each guillemot colony, and the relative importance of different prey throughout the study area. The final reports will include results of census efforts, reproductive success, prey species and indications of predation noted at the study sites, with recommendations for restoration efforts or enhancement of natural recovery.

VI. SCHEDULES AND PLANNING

A. Report Submission Schedules

March-April 1991	Prepare for field season hire personnel
May-August 1991	Conduct field work
September-November 1991	Data input and analysis
December 1991	Draft report completed
February 1992	Final report completed

B. Sample and Data Archival.

Original copies of field data will be archived in the U.S. Fish and Wildlife Service (Service) oil spill file system. Copies of the data set will be archived with the Service's Marine and Coastal Bird Project.

C. Management Plan.

The Study Leader for the Pigeon Guillemot portion of the study has not been selected. Brad Andres will serve as the study leader for black oystercatchers. They will both work under the direct supervision of the Project Leader, Marine and Coastal Bird Project, Division of Migratory Birds, Fish and Wildlife Service, Anchorage, Alaska. The co-study headers will be responsible for coordinating the completion of field data collection, analysis of field data, and timely reporting of the information in draft and final reports. The Project Leader is responsible for achieving coordination with all other marine bird studies during the planning, implementing, and reporting phases of the study.

D. Logistics.

To complete this study will require the use of a 25-foot vessel and field camps at Naked, Montague, Green, and Knight islands, and other appropriate locations in the Sound.

VII. BUDGET

Salaries

Study Leader (PiGu)	\$48,000
Study Leader (BLOY)	20,000
Biotech (Pi Gu)	9,100
Biotech (Pi Gu)	9,100
Biotech (BLOY)	<u>9,100</u>
	\$95,300
Travel/Per Diem	5,000
Supplies	5,000
Equipment	10,000
Contract (otolith analysis and mussel contaminant analysis)	<u>4,700</u>
Total	\$120,000

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PROPOSED OIL SPILL RESTORATION STUDY

I. Cover

Title: Identification of Upland Habitats Used by Marbled Murrelets in Prince William Sound

Study Identification Number: To be Assigned
Name of Study Leaders: Kathy Kuletz

Lead Agency: U.S. Fish & Wildlife Service
Marine and Coastal Bird Project
Anchorage, Alaska

Cooperating Agency: U.S. Forest Service
Glacier Ranger District
Girdwood, Alaska

Cost of Proposal: \$124,200

Inclusive Dates of Study Plan: March 1, 1991 - February 28, 1992

Signatures

Study Leader: _____

Supervisor: _____

Oil Spill Coordinator: _____

Financial Officer: _____

II. Introduction

The marbled murrelet (Brachyramphus marmoratus), a small nearshore alcid, is a species of concern from Alaska to California. They were listed as threatened in British Columbia in 1990 and are being considered for threatened or endangered status throughout its range in the United States. Loss of nesting habitat is postulated as the reason for their decline in B.C., Washington, Oregon and California. Population estimates for murrelets are not available for

all of Alaska, but the area affected by the oil spill is believed to be a population center in Alaska (Mendenhall 1988). Marbled murrelets suffered direct mortality from the Exxon Valdez oil spill disproportionate to their numbers at risk (Piatt et al. 1990). Preliminary analysis suggest a non-significant decline in summer populations in Prince William Sound (PWS), compared to pre-oil spill estimates. The winter population declined significantly along oiled shorelines compared to unoiled shorelines (Laing pers. comm.). In 1989, the Naked Island area showed population declines compared to pre-spill numbers, possibly due to human disturbance causing temporary displacement (Kuletz 1990a). Murrelets collected in the spill zone in August 1989, were exposed to petroleum hydrocarbon contaminants (Robinson-Wilson, pers. comm.), suggesting the possibility of long-term effects.

Recovery of the murrelet population could be enhanced by ensuring the availability of undisturbed nesting habitat. The Restoration Planning Work Group, building on expert and public input, identified protection of upland nesting habitats as one way to assist the natural recovery of species which depend on upland habitats for some stage of their life cycle. To fulfill this objective, specific information is needed on habitat requirements of the marbled murrelet.

Unlike most other seabirds, there are no conspicuous sites used by large numbers of nesting murrelets. Murrelets are secretive and widely-scattered (non-colonial) during their breeding season. In lower latitudes, the birds nest in coastal old-growth conifers (Marshall 1988, Nelson 1990, Quinland and Hughes 1990). In Southcentral Alaska nesting requirements are unknown. There are qualitative accounts of tree nesting but no nests have actually been found. However, several ground nests have been found, some of which could have been the closely related Kittlitz's murrelet (B. breverostris).

In 1990, a restoration feasibility pilot study investigated methods of studying upland use by marbled murrelets on Naked Island. Using information obtained in 1990, this proposal presents a study plan to assist in the identification of murrelet nesting habitat and specific areas of nesting activity in PWS.

III. Objectives

- A. Refine the censusing protocol for marbled murrelets at upland sites in Prince William Sound.
- B. Document tree nesting of marbled murrelets in Prince William Sound.
- C. Determine the presence and absence of marbled murrelet in selected upland habitat sites in Prince William Sound.
- D. Describe habitat associations in documented use areas in Prince William Sound.

IV. Methods

Objective A: Refining censusing protocol for murrelets in Alaska

Through all aspects of this study, information will be collected that will help establish guidelines for conducting upland habitat surveys of nesting murrelets. The influence of weather, seasonal patterns and observational techniques will be considered. The 1990 feasibility project at Naked Island (Kuletz 1990b) occurred from 9 June to 18 August. The standard survey is done by field personnel conducting a "dawn-watch survey." To determine variability in detections and seasonal patterns, three of the stations which were surveyed at least three times in 1989 will be surveyed at least bi-monthly in 1991. In 1990, three types of the "dawn-watch" survey developed by the Pacific Seabird Group (Paton et al. 1989) were attempted. The "intensive" dawn-watch survey (observer remains in one location) proved most suitable for the remote, uneven terrain of Naked Island and will be the basic field method of determining upland murrelet activity in 1991.

A dawn-watch survey is done at a pre-selected site during peak murrelet activity, when birds fly to their nests to exchange incubation duties or feed chicks. Since Naked Island birds displayed the same pattern as those at lower latitudes, each survey will be 45 minutes before to 75 minutes after official sunrise. Weather and lighting conditions (using a photography light meter) will be noted. Observers will use a tape recorder to note time of observation, type of detection (audio, visual or both), number of birds, number and types of vocalizations, direction and distance from the observed, and murrelet behavior (flight patterns, height of bird). Because birds may pass over an area without nesting there, certain behavioral activities and height of the bird will be used to classify the station as a "documented use area" (Nelson 1990). Birds flying silently through or circling below tree canopy, landing in trees or making stationary calls from trees indicate a documented use area.

All observers will be trained prior to the surveys, particularly in the classification of "detections" and in identification of murrelet calls and flight patterns. Field personnel will receive training via videos and audio tapes in the Regional Office, and in the field with the Study Leader. Training could begin as early as April, using sites in Kachemak Bay. Training at the Naked Island sites will not begin in early or mid-May.

A pilot study will be implemented to test the efficacy of self-activated tape records in determining murrelet activity in upland areas. If operable under Alaskan conditions, this system would enable greater coverage of areas where the number of field personnel are limited and access is difficult. The tape recorder will be set to record during the period of a dawn-watch survey in conjunction with a field observer. Test surveys will be made with the recorder at different heights, in clearings and in the trees. Data from field observers and the tape recorder will be compared for similarities in the number of audio visual detections.

Objective B: Documentation of tree nests in southcentral Alaska

In 1990, several sites with high murrelet activity were located on Naked Island. In some cases, potential nest sites were narrowed down to a few trees and birds were observed to land in trees. Now that these areas are known, a major effort to locate nests will be made on Naked Island using the proven "intensive ground survey" method (Naslund 1990, Singer et al. in press). Multiple observers (2-3), connected by hand-held radios, will focus on specific clusters of trees, and eventually individual trees, to locate a potential nesting branch. Once a suspected nest branch is located, a tree climber will climb an adjoining tree to document the nest. Data will be taken on nests following the Pacific Seabird Groups Nest Site Sampling Protocol (Varoujean and Carter 1989).

The search for nests will be augmented by use of audio equipment which can detect the soft calls made at the nest by adults and juveniles, and the wing beats of birds landing in trees (Singer, pers. comm., Nelson, pers. comm.). A portable cassette tape recorder, equipped with headphones and parabolic reflector will be used at documented use sites or suspected nest areas.

Objective C: Determine presence and absence in selected sites

The relationship between at-sea counts of murrelets and their upland nesting areas is unknown. This study will test for a correlation between at-sea densities and upland activity on a coarse scale. Results will indicate if at-sea counts are a reliable indicator of nearby upland nesting by murrelets. If so, future efforts to locate "documented use areas" can be more readily focused.

The presence and absence of marbled murrelets will be determined using intensive dawn-watch surveys. Based on results of boat surveys of waterbirds in PWS (Laing, unpubl. data), two types of shoreline sections, those with high murrelet densities and those with low murrelet densities, will be selected as focal points for dawn-survey stations. A survey station will be established approximately 200 meters inland from the middle of the shoreline transect. Data from Naked Island in 1990 indicated that numbers and flight direction relative to major habitat features could be determined. Among the seven paired stations in 1990 (each with a site near shoreline and another further inland), the amount of murrelet activity near the water was correlated with activity further inland (Kuletz 1990).

A minimum of 10 inland survey sites will be selected in both high and low density areas. Each site will be visited three times between May and mid-August, with surveys separated by at least two weeks at a given site (Nelson, pers. comm.). Field personnel will conduct dawn-watch surveys as described in Objective A, such that pertinent data on local habitat-murrelet associations will be available.

These surveys will require a mobile crew of at least two people, supported by a 25-foot vessel for transport around the PWS. Allowing for weather days and logistics, one crew could complete the 60 surveys required for the minimum number of survey sites. Increasing the sample size will require a second crew of two observers, which could be supported by the same boat.

Objective D: Describe habitat associations in documented use areas

Because Naked Island has known use sites, efforts to describe plant associations on a fine scale will be conducted by the U.S. Forest Service (USFS). At documented nesting sites already mapped, a plant-association crew will conduct ground surveys to provide detailed habitat data. The USFS has received \$40,000 to conduct this aspect of the study. This effort could be expanded with a second two-person dawn-watch survey crew which would be assigned pre-selected survey sites throughout Naked Island. The survey sites would be selected randomly within four habitat types defined by analysis of aerial photographs. The USFS will also provide maps of timber types occurring on Naked Island and other sites in the PWS.

V. Data Analysis

A. Tests

Data collected during 1991 will be combined with data collected in 1989 and 1990 and analyzed using standard statistical protocols.

B. Products.

This study will provide maps, computerized data sets and a final report on marbled murrelet activity at all surveyed sites. Detailed data on habitat and timber types will be compiled for all documented use sites and nest sites, through the cooperation of the USFS. The presence and absence of murrelets will be correlated with habitat. These data can be used in subsequent phases of the study to test predictions of murrelet presence in the field.

VI. Schedules and Planning

A. Report Submission Schedule:

March-April 1991	Prepare for field season/hire personnel
May-August 1991	Conduct field work

Sept.-Nov. 1991	Data input and analysis
December 1991	Draft report completed
February 1992	Final report completed

B. Sample and Data Archival.

Original copies of field data will be archived in the USFWS oil spill file system. Copies of the data set will be archived with the USFWS marine and Coastal Bird Project and the USFS Glacier Ranger District.

C. Management Plan

Kathy Kuletz will serve as the Study Leader or principal investigator. Ms. Kuletz works under the direct supervision of the Project Leader, Marine and Coastal Bird Project, Division of Migratory Bird Management, Fish and Wildlife Service, Anchorage, Alaska. The Study Leader is responsible for coordinating the completion of field data collection (including the habitat association information, analysis of field data; and timely reporting of the information in draft and final reports. The Project Leader is responsible for achieving coordination with all other marine bird studies during the planning, implementing, and reporting phases of the study. The USFS investigators are responsible for completing the habitat association descriptions and timber typing as described in this proposal. The USFS investigators work under the general direction of the USFWS Study Leader, all of whom will cooperate toward the accomplishment of the study objectives.

D. Logistics

To complete this study will require the use of a 25-foot vessel and field camps in Cabin Bay on Naked Island and other appropriate locations in the PWS.

VII. Budget

Salaries and Overtime

Study Leader GS-11 (1FTE)	-	\$ 48,000
Biotech GS-7 (1FTE)	-	35,000
Biotech GS-5 (.4FTE)	-	9,100
Biotech GS-5 (.4FTE)	-	<u>9,100</u>
Total Salaries		\$101,200

Travel/Per Diem	-	\$ 7,000
Supplies	-	6,000
Equipment	-	<u>10,000</u>
Total		\$124,200

IX. CITATIONS

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RPWG
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MEMORANDUM

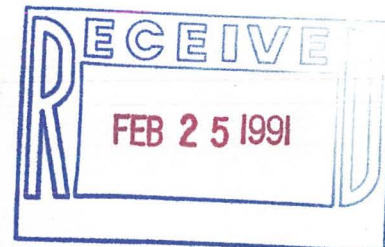
STATE OF ALASKA
DEPARTMENT OF FISH AND GAME

TO: Stan Senner
Restoration Planning Work Group
437 E Street
Anchorage, Ak. 99501

DATE: February 23, 1991

FROM: Daniel Sharp *DS*
Fisheries Biologist II
OSIAR Division
Cordova

PHONE: 424-3214



Subject: Detailed Study Plans for Coded Wire Tagging in 1991.

The following study plan is being submitted to the Restoration Planning Work Group. It contains the goals of the coded-wire tag application project for the period from March 1, 1991 to February 28, 1992. The contents of the study plan are derived from sections of the NRDA study plan for Fish/Shellfish Study 3 because this project is an extension of that study. The guidelines for the detailed study plans did not contain a sample budget format however detailed budget information is available in the original format prepared for the NRDA study plan. This study plan focuses only on the goals of the tag application portion of the comprehensive coded wire tagging studies. Tag recoveries from adult and juvenile salmon are performed as part of the NRDA efforts and their objectives are not included here. If greater detail or information is needed for your evaluation of any part of this study plan please inform me directly. Distribution of your letter dated the sixth of February took over one week to reach Cordova from Anchorage which made the deadline somewhat impractical during a rather busy time of the year. Thanks.

OBJECTIVES

1. Provide marked hatchery salmon of known origin and oil exposure history for recovery as adults to estimate catch and survival rates.
2. Provide marked wild pink salmon from three streams with contaminated estuaries and three streams with uncontaminated estuaries to estimate survival using outmigration, catch and escapement (using stream weirs).
3. Provide marked salmon of known origin and oil exposure history for recovery by researchers studying early marine migration, growth, and survival (F/S Study 4).
4. Provide marked wild sockeye salmon from two watersheds with contaminated estuaries and one watershed with an uncontaminated estuary to estimate survival using outmigration, catch and escapement (using stream weirs).
5. Continue to identify relevant injuries for which methods of restoring lost use, populations, and habitat must be developed.

METHODS

A subsample of fry or smolt from all hatcheries releasing salmon into PWS will be tagged with a coded wire tag (Appendix A). Wild stock pink fry and sockeye smolt from both oiled and non-oiled areas of the Sound will also be tagged (Appendix B). Tags will be applied at rates which will insure that, given a realistic recovery effort, sufficient numbers can be recovered in the commercial fishery, hatchery cost recovery harvests, and hatchery brood stock collections (Appendixes) to allow researchers to estimate the contribution of each tag release group by district, week, and processor stratum. Release groups represent differences in release timing or treatment (i.e. fed vs. unfed fry).

Tag application will be similar among all hatcheries and among all wild stock systems. Fry or smolt will be randomly selected as they emerge from incubators or raceways in hatcheries or outmigrate from streams. Selected fry will be anesthetized in a 1 ppm solution of MS-222, adipose fin clipped, and tagged. A random sample of 100 fish will be graded for fin clip quality each day. The proportion of bad clips in the sample will be used to discount the daily release of tagged fish. Clipped fish will be tagged and passed through a quality control device to test for tag retention. Fish repeatedly rejected will be killed to minimize the number of untagged but clipped fish in the release. Fish that retain tags will be held for 24 hours to determine short term mortality. A sample of tagged fish from each tagger will be taken each day and graded for tag placement according to

criteria developed by Peltz and Miller (1988). Prior to release, a 200 fish sample will be randomly sampled to estimate overnight tag retention. The proportion of lost tags in the sample will be used to estimate tag retention in the daily release. A written description of the tagging will be developed which will include a detailed description of each tag lot, the number of fish tagged, the total number of fish in the release lot, the average size of the fish at release, a profile of the exposure history of the release lot to the oil spill, and all information required by the ADF&G Coded-Wire Tag Laboratory which coordinated tagging in Alaska.

Release and tagging procedures are similar for pink and chum salmon. Both species are tagged with half length tags. At the Prince William Sound Aquaculture Corporation (PWSAC) hatcheries, tagged fry are released directly into large saltwater rearing pens with untagged fish of the same release group. At the Valdez Fisheries Development Association (VFDA) Solomon Gulch Hatchery tagged fry are placed in small enclosures within larger saltwater rearing pens for at least three days to allow them to recover from tagging before being mixed with unmarked fry from the same release group. At PWSAC hatcheries, unmarked fry entering large pens are counted with Northwest Marine Technology counters. At VFDA, unmarked fry in each pen are estimated from counts of eggs in incubators minus egg mortalities. At all facilities, mortalities in the large pens are estimated visually prior to release. Mortality rates based on visual estimates will be applied equally to tagged and untagged fish. The total numbers of fish in group t with valid tags at the time of release are estimated as

$$T_t = (T_t - M_t) - (T_t - M_t)L_t,$$

where

T_t = total number of fish tagged from group t,
 M_t = overnight mortality among fish tagged from treatment group t,
 L_t = overnight tag loss among fish tagged in treatment group t.

The VFDA estimate includes a term for short term mortality of tagged fish from treatment group t during saltwater rearing (S_t). The number of tagged fish released becomes

$$T_t = (T_t - M_t - S_t) - (T_t - M_t - S_t)L_t.$$

Hatcheries release fry when plankton monitoring indices indicate peak zooplankton abundance.

Four hatcheries released 615 million pink fry in 1990. Each of 32 release groups were tagged at a rate of approximately one tag per 580 fish released (1 in 500). The tag rate was held constant across release groups to prevent confusion of differential tag mortality with variation in survival between release groups (Peltz and Geiger, 1988; Geiger and Sharr, 1989). Release sizes and tagging rates will be approximately the same in 1991 but the number of release groups will increase (Appendix A).

In 1989, chum salmon were tagged at the rate of approximately one tag per 60 fish released at the Solomon Gulch Hatchery near Valdez. Tagging of Solomon Gulch chum salmon releases continued at the same level of effort at Solomon Gulch in 1990 and the WHN hatchery release of 20.6 million chum salmon fry was also tagged at a rate of approximately one tag per 480 fish released. The 1.6 million chum fry scheduled for release at Solomon Gulch in 1991 will be in two treatment groups each to be tagged at a rate of approximately 1 in . The scheduled 78 million chum fry release from WHN Hatchery will in four treatment groups, each tagged at a rate of approximately 1 in 500 (Appendix A).

Wild pink salmon were tagged from six stocks examined in F/S Study 2 in 1990; three from oil contaminated streams and three from streams which were not contaminated. Incline plane traps are used to capture fry as they emerge. Trapped fry were manually enumerated in 1990. Manual enumeration will continue in 1991 but electronic fry counters will also be tested. A portion of the daily outmigration are anesthetized and tagged. The anesthesia and associated trauma require that the tagged fish be held separate from their untagged cohorts, until they appear to have fully recovered from the effects of tagging. The extent to which the survival and behavior of the tagged fish can be extrapolated to other groups of salmon will be assessed at the time of recovery. Approximately 40,000 fry were tagged for each stock at tagging rates ranging from 1 in 4 to 1 in 17 fish released. Tagging will continue for these same stocks and at similar levels in 1991.

Because of they have similar freshwater rearing requirements, tagging procedures for hatchery stocks of sockeye, coho and chinook salmon smolt are similar. Full length tags are used for all three species. At each hatchery, a sample of smolt are captured from rearing appliances with nets in approximate proportion to the number of fish in the appliance. They are anesthetized, their adipose fin excised, and a tagged. A sample of fish from each day's tag production is retained to estimate short-term tag loss and tag induced mortality. Following tagging, the tagged fish are returned to mix with untagged cohorts. All mortalities during the first week after tagging will be examined and the tag status noted.

Smolt in the 2.6 million fish release of sockeye salmon from the

Main Bay Hatchery were tagged at a rate of 1 in 21 in 1990. The projected 2.6 million sockeye salmon smolt release from Main Bay in 1991 will be divided into 8 treatment groups which will each be tagged at a rate of approximately 1 in 22 (Appendix A). The majority of 2.3 million coho smolt scheduled for release from the WHN Hatchery in 1991 will be two groups to be released at the hatchery site. The remainder of the smolt will be apportioned between two groups destined for remote release sites. The two large releases at the hatchery site will be tagged with separate tag lots at a rate of approximately 1 in 40. The remote releases will be tagged separately at much higher rates. The scheduled 1991 release of 1.6 million coho smolt from Solomon Gulch Hatchery will occur in two groups which will be tagged with separate codes at a rate of approximately 1 in 80. The majority of the small release of chinook salmon smolt from WHN Hatchery will be tagged at a rate of 1 in 20 and released at the hatchery site. A small remote release at Cordova will be tagged at more than double that rate.

Wild stocks of sockeye salmon are tagged during the volitional smolt outmigration. Smolt are captured in traps as they migrate to saltwater, anesthetized, and tagged. Full length tags are used for two of the wild stocks but half length tags are used for the small smolt from Coghill Lake. The anesthesia and associated trauma require that the tagged fish be held separate from their untagged cohorts until they appeared to have fully recovered from the effects of tagging. As in the wild pink salmon tagging, the extent to which the survival and behavior of the tagged fish can be extrapolated to other groups of salmon will be assessed at the time of recovery. Wild sockeye salmon smolt were tagged at Eshamy and Coghill Lakes in 1989 and at Eshamy and Jackpot Lakes in 1990. Tagging of sockeye salmon will continue all three streams in 1991. Approximately 30,000 smolt are scheduled to be tagged from each of the three stocks but the actual number tagged and the tagging rates will be dependent upon the size and rate of outmigration.

DATA ANALYSIS

Data analysis will take place over many years as tagged salmon continue to return as adults. This years coded-wire tagging of salmon fry will result in tagged adults returning as soon as 1992. The analysis of 1989 through 1991's tag return data is funded as part of the 1991 NRDA study plan.

Tag recoveries are expected from releases at all four pink salmon hatcheries in 1990. Recoveries are expected for chum salmon from Main Bay Hatchery in 1986, Main Bay and Solomon Gulch Hatcheries in 1987, and Solomon Gulch in 1989. Tagged sockeye salmon will be recovered from the Main Bay facility releases in 1988 and 1989, and the Eshamy wild stock tagging of 1989. Tagged coho salmon will be recovered from 1990 releases at Wallace H. Noeremberg (WHN) and Solomon Gulch Hatcheries.

The recovery samples are from a stratified sample (Cochran 1977), by district and discrete time segments. The recovery will be further stratified by processor as described in Peltz and Geiger (1988). For each time and area specific stratum, 15% of the pink salmon catch and a minimum of 20% of other salmon species catches will be scanned for fish with a missing adipose fin. Catch sampling will be done in four fish processing facilities in Cordova, one facility in Seward, and three facilities in Valdez. When feasible, sampling will occur at facilities in Kodiak, Kenai, Anchorage, and Whittier and on large floating processors. 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.

In addition to catch sampling at the processing facilities, approximately 15% of the fish in the hatchery terminal harvest areas will be scanned for fish missing adipose fins. There will be a brood stock tag recovery effort at each of the three hatchery facilities where tags were initially applied. A minimum of 50% of the daily brood stock requirements of each facility will be scanned for fish with missing adipose fins.

Tags from wild stocks of sockeye salmon and pink salmon will be recovered coincidental with recoveries of hatchery stocks in the commercial catch, terminal harvest, and brood stock sampling programs. Tags for will also be recovered in the escapements of each tagged wild stock. At each of these streams crews will enumerate the daily escapement through a weir. At sockeye salmon weirs, a portion of the escapement passed through the weir each day will be scanned for missing adipose fins. At pink salmon weirs, daily foot surveys of each stream will be conducted to enumerate fresh carcasses and scan them for missing adipose fins. Carcasses enumerated each day will be marked to prevent duplicate counting on subsequent days and heads will be collected from all carcasses with adipose fin clips.

In the catch, terminal harvest, brood stock, and natural system surveys, the total number of fish scanned and the total number of fish with missing adipose fin will be recorded. The heads will be removed from fish with missing adipose fins. Each head will be tagged with uniquely numbered strap tags. Recovered heads will be assembled and pre-processed in the Cordova area office. Heads will then be sent to the FRED Division Coded-Wire Tag Laboratory in Juneau for decoding and data posting.

A statewide coded-wire tag lab is located in Juneau and operated by FRED Division of ADF&G. Coded-wire tag sampling forms will be checked for accuracy and completeness. Sampling and biological data will first be entered onto the laboratory's data base. Next, the heads will be processed. This involves removing and decoding the tags, and entering the tag code and the code assigned in the recovery survey into the database. Samples will be processed within five working days of receipt.

The first step in the coded-wire tag analysis will be to estimate the harvest of salmon from each tag lot, in units of adult salmon. Adult salmon from these tagged lots will be recovered in the common property fishery, the hatchery cost recovery fishery, and the adult brood stock. For the hatchery stock, a modification of the methods described in an ADF&G technical report by Clark and Bernard (1987) will be used. The specific methods, estimators, and confidence interval estimators are described in ADF&G technical reports on two previous studies of pink salmon in PWS: Peltz and Geiger (1988), and Geiger and Sharr (1989). Additional references on methods of tagging pink salmon in PWS can be found in Peltz and Miller (1988). In the case of the wild stocks, the methods and estimators and necessary assumptions are described by Geiger (1988).

The contribution of a particular tag lot, to a particular fishery stratum, is estimated multiplying by the number of tags recovered in the structured recovery survey, by both the inverse of the proportion of the catch sampled (the inverse sampling rate), and by the inverse of the proportion of the tag lot that was actually tagged (the inverse tag rate). The escapement (brood stock) of each tag lot is estimated using methods unique to the particular situation. After the contribution to each fishery is estimated for the tag lot, the survival is estimated by summing the estimated harvest of the tag lot in each fishery, and the estimated escapement (brood stock), and dividing by the estimated number of fish represented by the tag code.

Total catches stratified by week, district, and processor were obtained from summaries of fish sales receipts (fish tickets) issued to each fisherman. The total hatchery contribution to the commercial and hatchery cost recovery harvest is the sum of the estimates of contributions in all week, district, and processor strata:

$$\hat{C}_t = \sum_i X_{ti} (N_i / S_i) p_t^{-1}$$

where:
 \hat{C}_t = catch of group t fish,
 X_{ti} = number of group t tags recovered in i th strata,
 N_i = number of fish caught in i th strata,
 S_i = number of fish sampled in i th strata,
 p_t = proportion of group t tagged.

For sampled strata, we used a variance approximation which ignores covariance between release groups (Geiger 1988):

$$V(\hat{C}_i) = \sum X_{ti} (N_i/S_i) p_i^2 [1 - (N_i/S_i) p_i^{-1}].$$

The average tag recovery rate for all processors in a week and district will be used to estimate hatchery contribution in catches delivered to processors not sampled for that district and week. Variances associated with unsampled strata will not be calculated.

SCHEDULES AND PLANNING

Appendix A lists the 1991 tagging goals for all hatchery fish and Appendix B lists the goals for wild fish. Tagging at some locations will begin as soon as March 1, 1991 and will be complete by June 1, 1991 at all locations. Finalized tagging and release data will be available on July 1, 1991.

Summary Table

March 1	Tagging begins at PWSAC and VFDA.
March 17.	Wild Pink salmon tagging camps erected.
April 1	Tagging begins at Pink salmon camps.
May 1	Tagging begins at Sockeye salmon camps.
June 1	Tagging complete at all hatcheries.
June 15	Tagging of wild salmon complete.
	Finalized tagging data due from PWSAC and VFDA. Adult sockeye weirs erected at Eshamy and Jackpot for tag recovery.
July 1	Finalized tagging data sent to Tag Lab.
September 10.	Adult sockeye weirs removed.
November 1	Prepare Preliminary Restoration Status Report.
November 15	Submit order for coded wire tags and equipment for 1992.
December 15	Prepare project operational plans for 1992.

Data Archival

This restoration tagging study is a continuation of a damage assessment coded wire tag study. The objectives of the tag application portion of these studies does not generally require the handling of hydrocarbon samples or samples that require chain of custody procedures. The tag application portion primarily generates data that is treated as confidential by all parties involved with the tag applications. Field notes and data records are kept and copies of reports and forms that contain results are stored in the Cordova office of ADF&G.

Management Plan

All coded wire tagging in Prince William Sound will be coordinated by the Tagging Project Leader, a Fisheries Biologist II in the Cordova office of ADF&G. The Project Leader will arrange tagging contracts and coordinate tagging schedules for the PNP hatchery operators and the Department of Fish and Game. The Department of Fish and Game's tag application efforts employ roughly 26 people at the peak of the tagging season. The Project Leader is assisted by two Fisheries Biologist I's, seven Fish and Wildlife Technician III's and sixteen Technician II's.

Logistics

Field camps for this project were established in 1989 and 1990. Equipment was purchased as part of the NRDA study in 1989. Leases and permits have been secured from the proper authorities and arrangements have been made to initiate field activities in mid-March. Field work will continue into September for some components of the project. Personnel in the tagging program have two years experience at operating these same remote field camps and have been preparing for the field season since December.

BUDGET: ADF&G

Salaries	\$425.0
Travel	2.5
Contracts	322.0
Supplies	63.0
Equipment	109.0
Total	\$922.0

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Appendix A. Coded-wire tagging goals for hatchery releases of salmon in PWS, 1991.

Hatchery	Projected Species	Tag Release	Valid Tags to Goal	Number Ratio Order	Total Release /Marked Number of Goal	Tag Codes	Length
Armin F. Koernig	Pink	116,000,000	193,000	218,000	600	16	Half
Cannery Creek	Pink	140,000,000	234,000	261,000	600	14	Half
Solomon Gulch	Pink	140,000,000	233,000	252,000	600	10	Half
Wally Norenburg	Pink	225,000,000	375,000	422,000	600	18	Half
GRAND TOTAL	Pink	621,000,000	1,035,000	1,153,000	600	58	Half
Solomon Gulch	Chum	1,600,000	20,000	20,000	80	2	Half
Wally Norenburg	Chum	76,000,000	156,000	173,000	500	4	Half
GRAND TOTAL	Chum	79,600,000	176,000	193,000	450	6	Half
Solomon Gulch	Coho	1,000,000 20,000	30,000 10,000	30,000 10,000	33 2	2 1	Full Full
Wally Norenburg	Coho	2,300,000	73,500	73,500	40	2	Full
Whittier	Coho	100,000	10,000	20,000	10	1	Full
Cordova	Coho	50,000	10,000	10,000	5	1	Full
GRAND TOTAL	Coho	3,470,000	133,500	143,500	26	7	Full
Main Bay	Sockeye	3,575,000	125,000	125,000	29	8	Full
GRAND TOTAL	Sockeye	3,575,000	125,000	125,000	29	8	Full
Wally Norenburg	King	600,000	30,000	30,000	20	1	Full
Cordova	King	60,000	10,000	10,000	6	1	Full
GRAND TOTAL	King	660,000	40,000	40,000	17	2	Full
GRAND TOTAL	All	708,305,000	1,509,500	1,654,500	470	81	Both

Appendix B. Coded-wire tagging goals for wild stocks of salmon in PWS, 1991.

System	Treatment	Species	Projected Outmigration	Valid Tag Goal	Total Release /Marked Ratio	Number of Tag Codes	Tag Length
Upper Herring B.	Oiled	Pink	210,000	40,500	5	3	Half
Hayden Ck.	Oiled	Pink	360,000	40,500	9	3	Half
Loomis Ck.	Oiled	Pink	210,000	40,500	5	3	Half
Cathead Ck.	Clean	Pink	150,000	40,500	5	3	Half
O'Brien Ck.	Clean	Pink	300,000	40,500	7	3	Half
Totemoff Ck.	Clean	Pink	720,000	40,500	18	3	Half
GRAND TOTAL	All	Pink	1,950,000	243,000	8	18	Half
Coghill	Clean	Sockeye	600,000	27,000	22	2	Half
Eshamy	Oiled	Sockeye	600,000	27,000	22	2	Full
Jackpot	Oiled	Sockeye	600,000	27,000	22	2	Full
GRAND TOTAL	All	Sockeye	1,800,000	81,000	22	6	Both
GRAND TOTAL	All	All	3,750,000	323,000	30	24	Both

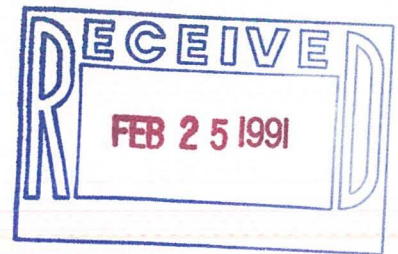
Appendix A. Coded-wire tagging goals for hatchery releases of salmon in PWS, 1991.

Hatchery	Projected Species	Tag Release	Valid Tags to Goal	Number Ratio Order	Total Release /Marked Number of Goal	Tag Codes	Length
Armin F. Koernig	Pink	116,000,000	193,000	218,000	600	16	Half
Cannery Creek	Pink	140,000,000	234,000	261,000	600	14	Half
Solomon Gulch	Pink	140,000,000	233,000	252,000	600	10	Half
Wally Norenburg	Pink	225,000,000	375,000	422,000	600	18	Half
GRAND TOTAL	Pink	621,000,000	1,035,000	1,153,000	600	58	Half
Solomon Gulch	Chum	1,600,000	20,000	20,000	80	2	Half
Wally Norenburg	Chum	78,000,000	156,000	173,000	500	4	Half
GRAND TOTAL	Chum	79,600,000	176,000	193,000	450	6	Half
Solomon Gulch	Coho	1,000,000	30,000	30,000	33	2	Full
		20,000	10,000	10,000	2	1	Full
Wally Norenburg	Coho	2,300,000	73,500	73,500	40	2	Full
Whittier	Coho	100,000	10,000	20,000	10	1	Full
Cordova	Coho	50,000	10,000	10,000	5	1	Full
GRAND TOTAL	Coho	3,470,000	133,500	143,500	26	7	Full
Main Bay	Sockeye	3,575,000	125,000	125,000	29	8	Full
GRAND TOTAL	Sockeye	3,575,000	125,000	125,000	29	8	Full
Wally Norenburg	King	600,000	30,000	30,000	20	1	Full
Cordova	King	60,000	10,000	10,000	6	1	Full
GRAND TOTAL	King	660,000	40,000	40,000	17	2	Full
GRAND TOTAL	All	708,305,000	1,509,500	1,654,500	470	81	Both

Appendix B. Coded-wire tagging goals for wild stocks of salmon in PWS, 1991.

System	Treatment	Species	Projected Outmigration	Valid Tag Goal	Total Release /Marked Ratio	Number of Tag Codes	Tag Length
Upper Herring B.	Oiled	Pink	210,000	40,500	5	3	Half
Heyden Ck.	Oiled	Pink	360,000	40,500	9	3	Half
Lcomis Ck.	Oiled	Pink	210,000	40,500	5	3	Half
Cathead Ck.	Clean	Pink	150,000	40,500	5	3	Half
O'Brien Ck.	Clean	Pink	300,000	40,500	7	3	Half
Totemoff Ck.	Clean	Pink	720,000	40,500	18	3	Half
GRAND TOTAL	All	Pink	1,950,000	243,000	8	18	Half
Coghill	Clean	Sockeye	600,000	27,000	22	2	Half
Eshamy	Oiled	Sockeye	600,000	27,000	22	2	Full
Jackpot	Oiled	Sockeye	600,000	27,000	22	2	Full
GRAND TOTAL	All	Sockeye	1,800,000	81,000	22	6	Both
GRAND TOTAL	All	All	3,750,000	323,500	30	24	Both

Cetacean Monitoring Studies



Study ID Number: Marine Mammals Study Number 6

Project Leader(s): Marilyn E. Dahlheim and Thomas R. Loughlin

Lead Agency: National Oceanic and Atmospheric Administration

Cooperating Agency(ies): Federal: None
State: None

Cost of Proposal: \$140.0K

Dates of Study Plan: 1 April 1991 through 31 March 1992
(Year 1).

Marilyn E. Dahlheim, Ph.D.
Project Leader

Thomas R. Loughlin, Ph.D.
Project Leader

Howard W. Braham, Ph.D.
Organization Leader

Joanne Wejak
Financial Officer

Alaska Fisheries Science Center
National Marine Mammal Laboratory
7600 Sand Point Way N. E., Bin C15700
Seattle, Washington 98115-0070
206/526-4045

15 February 1991

INTRODUCTION

There are more than 25 species of marine mammals in Prince William Sound, the Gulf of Alaska, and adjacent waters. The area impacted by the Exxon Valdez oil spill (EVOS) provides a variety of marine habitats seasonally critical for significant numbers of these mammals. Damage assessment studies concentrated on four marine mammal species; harbor seals, Steller sea lions, killer whales, and humpback whales, principally because the historical data base on these species was adequate for comparative purposes and the ability for demonstration of injury was high. However, other species, such as the reclusive harbor porpoise and ubiquitous Dall's porpoise may have been significantly affected by the spill. Injury to marine mammals may have resulted in death, reduced fitness, or destruction or modification of habitat. The following proposed studies will measure the probable success of restoration plans to determine the long-term improvement in species recovery (measured by changes in vital rates and abundance trends) and restoration of that species habitat (measured by distribution, use, and density of marine mammals). Damage assessment studies have demonstrated changes in both these parameters for killer whales and harbor seals; similar data for sea lions and humpback whales are equivocal.

The proposed monitoring studies will provide additional information on the health and stability of cetacean populations within the Prince William Sound ecosystem. Cetaceans are high trophic level predators and their distribution, abundance, and vital rates are viable measures of the health and stability of the ecosystem. Changes in food availability, habitat degradation, and ecosystem stability can be inferred by reduced cetacean abundance, declining trends, or reduced reproduction.

OBJECTIVES

- A. To assess cetacean reproductive rates and trends in abundance within Prince William Sound and adjacent waters. Trend surveys will concentrate on killer whales, but other species will be monitored as appropriate.
- B. To assess critical habitat for killer whales and other cetacean species by monitoring distribution and density of each species in the study area.

METHODS

A. Sampling Methods

The objectives of the study will be addressed primarily by conducting aerial and shipboard surveys within Prince William Sound, in both oiled and unoled areas, and in areas outside

Prince William Sound from Cape St. Elias to the Kodiak Archipelago. The scope of monitoring will be similar to that conducted during the NRDA but will include additional surveys to assess abundance and habitat preferences of marine mammal species not evaluated by initial NRDA studies. Surveys will be designed to incorporate the collection of data on all marine mammal species occurring seasonally in the study area with emphasis on killer whales. Success of the study will be measured by the adequacy of the data to meet the objectives.

Cetacean Reproductive Rates/Trends in Abundance

To achieve Objective 1, vessel and aerial platforms will be used to collect information on cetacean abundance and distribution. Line or strip transect survey design will be employed to provide adequate coverage of the study area. Specific areas, known for cetacean concentrations, will be investigated during survey transects. If reports of whales are received from other sources (e.g, sighting network established in Prince William Sound), those areas will also be examined after completion of systematic surveys. All cetaceans observed during the surveys will be recorded.

Killer whale reproductive rates and trends in abundance will be investigated by using ship surveys to obtain photographs of individual whale identifications. The majority of killer whale photo-identification work performed during the 1991 season in Prince William Sound will be conducted under damage assessment studies (Marine Mammal Study Number 2 - Year 3). When killer whales are sighted, researchers will stop further search efforts (logging off effort) and approach the whales to collect photo-identification information. The photographic techniques used to obtain high-quality images of killer whales are described in detail in the damage assessment study plan. Reproductive rates and population trends of Prince William Sound humpback whales will also be investigated by collecting photographs of individual whales. Similar camera systems, film, and data collection techniques will be used while photographing humpback whales as those described above for killer whales.

In addition to photographs, data will be collected on the general conditioning of individual whales (i.e., observations of skin disease, measurement of respiratory cycles, etc). Stranded animals will be examined when possible.

Daily vessel effort logs are maintained each day which will permit 1) quantification of the amount of time searching for cetaceans vs photographing cetaceans, 2) quantification of search effort under different weather conditions; 3) projection of daily vessel trackline, and 4) the number of vessels/aircraft encountered in the study area. Cetacean density and distributional patterns within the study area will be evaluated

based on vessel surveys.

Aerial transects will be conducted following standard strip census procedures. Aerial surveys will take place both within Prince William Sound as well as in the immediate waters adjacent to Prince William Sound. Pre-defined tracklines will be flown at approximately 305 m (1000 feet) and an airspeed of 145km/h (90nm/h). The twin-engine, high-winged aircraft will be equipped with Loran C or GNS, and intercom. The research team will consist of at least three biologists. Aerial survey data will be used in conjunction with the vessel data to obtain estimates of cetacean density and distribution within the study area.

During the first year of investigations off Kodiak, we will rely primarily on the use of aerial surveys to obtain estimates of cetacean density and distribution. All existing cetacean data for the area will be reviewed, summarized and compared to the 1991 results. In subsequent years, depending upon the results of year one investigations, vessel surveys may be included into the Kodiak investigations. If vessel surveys are included in later years, they would follow the same design as those described above for the inside waters of Prince William Sound. Photographs of killer whales and humpback whales would be collected. Observations of all cetaceans would be recorded. Aerial survey data, vessel data, and all existing information would be used to evaluate cetacean density and distribution off Kodiak, Alaska.

Critical Habitat

Initial assesement of important habitat for killer whales and other cetacean species in Prince William Sound (Ojective 2) will include a description of marine mammal use areas characterizing location in relation to land, water depth, sea temperature, food, general behavior of animal in the area, and other animals present. Data pertinent to habitat evaluation will be collected during the vessel and aerial work described above. Review of pertinent fishery data will facilitate correlation of cetacean distribution with potential prey concentrations.

To properly evaluate important habitat, seasonal useage, and overall distributional patterns for killer whales, we will also determine the feasibility of placing satellite tags on Prince William Sound killer whales. Recently, many successful deployments of satellite telemetry packages on cetaceans and pinnipeds has provided important ecological information and each year brings about major advances in technology. If feasible and after extensive examination of existing technology, we will place satellite tags on at least three whales in Prince William Sound. Year one investigations (1991) will involve a review of all pertinent information pertaining to satellite tagging and a determination of the likelihood of success of placing satellite tags on killer whales. Scientists with expertise in satellite

tagging will be consulted. If the likelihood of success is considered high, year two (1992) would be devoted to the engineering of the satellite system for placement on killer whales. If necessary equipment is obtained in 1992 and we are satisfied with all aspects of the proposed work (delivery system, attachment device, minimal disturbance to whales, etc.), we would initiate satellite tagging during the 1993 season (year three).

DATA ANALYSIS

All exposed film of killer whales and humpback whales collected during the 1991 field season will be analyzed for individual identification. Details of photographic data analysis for both species is given in the damage assessment study plan. These individual whale identifications will permit an evaluation of species abundance, reproductive rates, and population trends.

To avoid biases in data interpretation, it is important that the amount of effort in searching for and photographing cetaceans during this monitoring study is at least equal to (but not less than) that completed in the damage assessment studies. When comparing differences in sightings per unit effort, either the Kolmogorov-Smirnov or Mann-Whitney test will be used. For a large killer whale pod (>12 animals), the likelihood of obtaining photographs of all individuals are increased as the number of encounters are increased. Some individuals, and certain pods, are more likely to approach vessels making photographic documentation easier; while others keep a considerable distance away making for more difficult conditions. Whale behavior also plays a role when attempting to obtain photographs of individual whales. If the pod is resting (typically grouped together), it is easier to obtain photographs of all whales vs when the pod is travelling (spread out through an area). Researchers with prior killer whale experience in a particular area, who are capable of recognizing individuals, will also enhance the likelihood of accounting for all whales within a pod.

Killer whale abundance, reproductive rates and trends will be determined by comparison to surveys conducted during NRDA and prior to the spill. Killer whale calves of the year will be noted and their mothers identified. Natality (number of calves per adult female) will be calculated for each pod for each year and comparisons made between resident and transient groups using descriptive statistics. Mortality rates will also be calculated for resident groups. Mortality for transient pods will be calculated when necessary data are available. General location of killer whales will be recorded each time photographs are taken, allowing comparisons of pod distributions among years.

Humpback whale abundance, reproductive rates, and trends will be determined by comparison to surveys conducted during NRDA and prior to the spill. Humpback calves will be noted and their

mothers identified. Natality (number of calves per adult female) will be calculated for each area. Comparisons of natality among years will be made using either Chi-square tests or Z tests for comparing differences between two proportions (selection of test based on sample size). Stranded animals will be reported. Distributional comparisons will be made on a qualitative basis.

Observations of cetaceans collected from both aerial and shipboard platforms throughout the season will be plotted and a chart of sightings generated. Abundance estimates for species other than killer whales and humpback whales will be calculated following standard line or strip transect procedures. Overall cetacean distributional patterns will be evaluated from the charts. A habitat summary will be provided to include information collected on water depth, water temperature, food, and relation of sighting location to land for all cetacean observations.

A progress report will be written assessing the feasibility of placement of satellite tags on killer whales based upon the information obtained during year one investigations. This would include recommendations for either continuation or cancellation of this aspect of the work.

SCHEDULES & PLANNING

A. Data Submission and Archival

A data submission schedule is attached listing milestone dates and activities (Attachment 1). No other special reports or additional visual data will be submitted other than those described in the reports.

Reports will be available through the National Marine Mammal Laboratory, Seattle, Washington (Attn: Drs. Dahlheim and Loughlin) summarizing the 1991 studies. Reports are written in a scientific format and contain an Abstract, Title Page, Table of Contents, List of Tables and Figures, Introduction, Materials and Methods, Results, Discussion, and Conclusion/Recommendation Section. Original survey forms, identification cards, daily logs, marine mammal sighting and effort forms are archived at the National Marine Mammal Laboratory. The highest quality photograph for each individual killer whale and humpback whale will be selected and a 2 1/2" by 3 1/2" print will be made for archival purposes.

All documents and materials associated with this monitoring study will be stored at the National Marine Mammal Laboratory, Seattle, Washington under the Alaska Ecosystem Program. Killer whale and humpback whale prints are stored in archival plastic sheets and properly labelled (date/location/photographer). Equipment

M I L E S T O N E C H A R T

SP #: Marine Mammals PI: Drs. Dahlheim & Loughlin

Study Number 6
Major Milestones: Reports, cruises, field effort, data management, workshops, significant contractual requirements, etc.

Actual Start Date ●
Planned Completion Date △
Actual Completion Date ▲

MAJOR MILESTONES	1991										1992			
	A	M	J	J	A	S	O	N	D	J	F	M	A	
Marine Mammal Organizational Meetings	▲													
PRINCE WILLIAM SOUND/ADJACENT WATERS														
Field Research: Photo-identification & vessel work		●	—			▲								
Aerial Surveys			●	—		▲								
Data Analysis & Draft Report		●	—							△				
Final Report & Products											△			
KODIAK ARCHIPELAGO														
Aerial Surveys			●	—		▲								
Data Analysis & Draft Report			●	—						△				
Final Report & Products											△			
SATELITTE TAGGING														
Basic Investigations	●	—								△				
Progress Report											△			

purchased for the investigations will be properly labelled. Serial numbers will be listed when available. Equipment will be stored in the custody of the Project Leaders at the NMML.

B. Management Plan

NOAA, Alaska Fisheries Science Center, National Marine Mammal Laboratory, 7600 Sand Point Way N. E., Bin C15700, Seattle, Washington 98115 (206/526-4045).

Dr. Marilyn E. Dahlheim, Project Leader

Duties: Project development, research design and implementation. Coordination of, and participation in, field research.

Dr. Thomas R. Loughlin, Project Leader

Duties: Project development and research design.

Ms. Joanne Wejak, Financial Officer

Duties: Administrative officer in-charge of processing financial paperwork associated with research.

Temporary Biologist

Duties: Laboratory/Field Assistant

NOAA, WASC, Procurement Division, 7600 Sand Point Way N. E., Bldg. 1, Location 22, Seattle, Washington 98115.

Duties: Contract Negotiations and Administration
206/526-6494

BUDGET**A. Costs (in thousands of dollars = K)**

	<u>Line</u>					<u>Total</u>
	<u>100</u>	<u>200</u>	<u>300</u>	<u>400</u>	<u>500</u>	
-Projected Expenses 4/91 - 3/92	45.0	9.0	80.0	2.0	4.0	\$140.0K*

* An additional \$186.0K is provided under Damage Assessment for Year One (1991). The annual budget of monitoring studies will be \$300.0K.

PROJECTED EXPENDITURE BREAKDOWN**Line 100 - Salaries**

<u>Level</u>	<u>Name</u>	<u>Months</u>	<u>Salaries & Benefits/Month</u>	<u>Total</u>
GM-14	Loughlin	1.0	5,800.00	5,800.00
GS-12	Dahlheim	5.0	4,200.00	21,000.00
GS-07	Assistant	6.0	2,275.00	13,650.00
GS-07	Wejak	0.3	2,400.00	800.00
GS-04	Assistant	3.0	1,275.00	3,750.00
			Total	\$45,000.00

Line 200 - Travel

*Research team of 3.

Seattle, Washington to Kodiak, Alaska & Return	2,100.00
Seattle, Washington to Prince William Sound, Alaska & Return	2,800.00
Per Diem (30 days)	4,100.00
Total	\$ 9,000.00

Line 300 - Contractual

Vessel/Aircraft contracts not awarded as of 15 February 1991.

Total \$80,000.00

Line 400 - SuppliesA. Field, film & processing
and marine supplies

2,000.00

Total \$ 2,000.00

Line 500 - Equipment

A. Computer hardware/software

4,000.00

Total \$ 4,000.00

GRAND TOTAL \$140,000.00

PERSONNEL QUALIFICATIONS

Curriculum Vitae of Project Leaders is provided (Attachment 2 and 3).

CURRICULUM VITAE (abbreviated)

Marilyn E. Dahlheim, Ph.D.
National Marine Mammal Laboratory
7600 Sand Point Way N. E., Bin C15700
Seattle, Washington 98115-0070

From 1978 to the present time have participated and designed marine mammal vessel and aerial surveys in Alaskan waters (Bering, Chukchi and North Pacific). Have collected and analyzed acoustical data on whales and seals inhabiting Arctic waters from vessel, ice, and helicopter platforms. Collected data on movements, behavior, and distribution of marine mammals and correlated distributional data on marine mammals with physical environment. Co-chief scientist on USCGC Icebreaker POLAR SEA in charge of shipboard activities and selection of personnel from multidisciplinary fields to define winter habitat of bowhead whales. Helped developed use of passive acoustics as a censusing device to monitor whales. Training of personnel on correct methods of collection and analysis of scientific data. Responsible for reviewing outside research proposals for accuracy of scientific hypotheses and methods. Review of numerous environmental assessments, impact statements, and marine mammal permits. Reviewer for two scientific journals and participation with other governmental agencies regarding solutions to problems arising from increasing oil development and vessel traffic and the acoustical effect on marine mammals. Principal investigator for five consecutive years conducting acoustical research on gray whales in Mexico. Principal investigator gray whale census (three consecutive years). Task leader on killer whale/blackcod fishery interactions in Prince William Sound, including photo-identification research. Task leader for photo-identification studies on killer whales in the Bering Sea (four years). Project leader on NRDA studies 1989-1991 on humpback and killer whales. Representative of the National Marine Mammal Laboratory at international conferences/ meetings; submission/acceptance of independent research proposals. Has published extensively in peer reviewed scientific journals and lay publications.

CURRICULUM VITAE (abbreviated)

Thomas R. Loughlin, Ph.D.
National Marine Mammal Laboratory
7600 Sand Point Way, NE
Seattle, WA 98115-0070

From 1977 to 1981 was Acting Chief, Research and Management Division, NMFS, Washington, D.C., and was responsible for development, implementation, and coordination of the national research and management program consisting of research into the life history and population dynamics of marine mammals and endangered species. Currently is leader of the Bering Sea/Gulf of Alaska Ecosystem Program, National Marine Mammal Laboratory and is responsible for developing and executing ecosystem based research regarding marine mammal abundance, distribution, trophic relationships, and environmental and fishery data throughout Alaska. Also responsible for the design, supervision, and execution of research addressing marine mammal fishery interactions between foreign and domestic commercial fisheries in Alaska. Has been Chief Scientist on numerous ship and terrestrial research programs spanning fifteen years of marine mammal research along the west coast of North America. Associate Professor (courtesy), Oregon State University, and reviewer for scientific papers submitted to over eleven scientific journals. Has published extensively in peer reviewed scientific journals and lay publications.

SELECTED CITATIONS

The following killer whale articles are pertinent to the studies.

- Anon. 1982. Report on the workshop on identity, structure, and vital rates of killer whale populations. Rept. Int. Whal. Commn, 32: 617-631..
- Balcomb, K. C. 1978. Orca Survey 1977. Final Report of a Field Photographic Study Conducted by the Moclips Cetological Society in Collaboration with the U. S. National Marine Fisheries Service on Killer Whales (Orcinus orca) in Puget Sound. Unpub. Report to the Marine Mammal Division, National Marine Fisheries Service, Seattle, Washington, 10 pages.
- Bigg, M. A. 1982. An Assessment of Killer Whale (Orcinus orca) Stocks off Vancouver Island, British Columbia. Rept. Int. Whal. Commn., 32: 655-666.
- Braham, H. W. and M. E. Dahlheim. 1982. Killer Whales in Alaska Documented in the Platforms of Opportunity Program. Rept. Int. Whal. Commn. 32: 643-646.
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- Ellis, G. 1987. Killer Whales of Prince William Sound and Southeast Alaska. A Catalogue of Individuals Photoidentified, 1976-1986. Sea World Research Institute/Hubbs Marine Research Center, Technical Report No. 87-200. April 1987.
- Fowler, C. W. 1984. Density Dependence in Cetacean Populations. In "Reproduction in Whales, Dolphins, and Porpoises". Eds. W. F. Perrin, R. L. Brownell, and D. P. DeMaster. Rept. Int. Whal. Commn., Spec. Issue 6: 373-380.
- Hall, J. D. 1981. Aspects of the Natural History of Cetaceans of Prince William Sound. Ph.D. Dissertation. University of California - Santa Cruz. 148 pp.
- Heyning, J. E. and M. E. Dahlheim. 1988. Orcinus orca. Mammalian Species Account, No. 304, pp. 1-9, 4 figs.
- Leatherwood, S., K. C. Balcomb, C. O. Matkin, and G. Ellis. 1984. Killer Whales (Orcinus orca) of Southern Alaska - Results of Field Research 1984 Preliminary Report. Hubbs Sea World Research Institute Tech. Report No. 84-175, 59 pp.
- Leatherwood, S., A. Bowles, E. Krygier, J. D. Hall, and S.

- Ignell. 1985. Killer Whales (Orcinus orca) in Southeast Alaska, Prince William Sound, and Shelikof Strait; A Review of Available Information. Rept. Int. Whal. Commn., SC/35/SM 7., 10 pp.
- Perrin, W. F. and S. B. Reilly. 1984. Reproductive Parameters of Dolphins and Small Whales of the family Delphinidae. In "Reproduction in Whales, Dolphins, and Porpoises". Eds. W. F. Perrin, R. L. Brownell, and D. P. DeMaster. Rept. Int. Whal. Commn., Spec. Issue 6: 97-134.
- von Ziegesar, O., G. Ellis, C. Matkin, and B. Goodwin. 1986. Repeated Sightings of Identifiable Killer Whales (Orcinus orca) in Prince William Sound, Alaska 1977-1983. Cetus, Vol. 6, No. 2, 5 pp.

The following humpback whale articles are pertinent to the studies.

- Baker, C. S. 1985. The Population Structure and Social Organization of Humpback Whales (Megaptera novaeangliae) in the Central and Eastern North Pacific. Ph.D. Dissertation. University of Hawaii. 306 pp.
- Baker, C. S. and L. Herman. 1987. Alternative population estimates of Humpback Whales (Megaptera novaeangliae) in Hawaiian Waters. Canadian Journal of Zoology, 65: 2818-2821.
- Baker, C. S., S. R. Palumbi, R. H. Lambertsen, M. T. Weinrich, J. Calambokidis, and S. J. O'Brien. 1990. Influence of seasonal migration on geographic distribution of mitochondrial DNA haplotypes in humpback whales. Nature 344:238-240.
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- Johnson, J. J. and A. A. Wolman. 1984. The Humpback Whale (Megaptera novaeangliae). Marine Fisheries Review 46(4):30-37.

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- Watkins, W. A., K. E. Moore, D. Wartzok, and J. H. Johnson. 1981. Radio Tracking of Finback (Balaenoptera physalus) and Humpback (Megaptera novaeangliae) Whales in Prince William Sound, Alaska. Deep-Sea Research 78: 577-588.
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- von Ziegesar, O. 1984. A Survey of the Humpback Whales in Southwestern Prince William Sound, Alaska 1980, 1981, and 1983. A Report to the State of Alaska, Alaska Council on Science and Technology, 68 pp.
- von Ziegesar, O. and C. O. Matkin. 1989. A Catalogue of Prince William Sound Humpback Whales Identified by Fluke Photographs Between the Years 1977 and 1988. 28 Pages. North Gulf Oceanic Society, P. O. Box 15244, Homer, Alaska

N1751
N

rec'd - RPO
3/1/91

MEMORANDUM

TO: Stan Senner, John Strand, Linda Comerici, USEPA Oil Spill
Restoration Planning Office, Anchorage, AK

FROM: Hal Kibby, Branch Chief, Ecotoxicology, USEPA ERL-C AF for HJK

SUB: PWS/GOA Tidal Marsh Restoration Feasibility Proposal

As per your letter of February 6, 1991, please find attached the research plan and QA/QC proposal for the "Feasibility of Restoring the Bay of Isles and Tonsina Bay in Prince William Sound and the Gulf of Alaska.

For your information the following individuals have reviewed drafts of the research plan and submitted oral or written comments:

- Hal Kibby, Branch Chief, Ecotoxicology, USEPA ERL-C
- Bill Williams, Former Team Leader, Ecotoxicology, ERL-C
- Anne Fairbrother, Team Leader, Ecotoxicology, ERL-C
- Dave Schmedding, MAN TECH ENVIRONMENTAL, ERL-C
- James Wyant, MAN TECH ENVIRONMENTAL, ERL-C
- Richard Meganck, MAN TECH ENVIRONMENTAL, ERL-C
- Richard Novitzki, MAN TECH ENVIRONMENTAL, ERL-C
- Ron Thom, Battelle NW
- Jay McKendrick, U. of Alaska
- Mel Knapp, Technical Resources Inc.
- Lori Suit, Technical Resources Inc.
- Brian Ross, USEPA
- Eric Preston, USEPA, ERL-C
- Ann Hairston, MAN TECH, ERL-C (editor)

The QA/QC document was prepared by Deborah Coffey, QA/QC Officer, MAN TECH ENVIRONMENTAL, ERL-C and reviewed by Meganck, Wyant, Thom McKendrick, Schmedding - above.

A. Title: FEASIBILITY OF RESTORING THE BAY OF ISLES AND TONSINA BAY IN PRINCE WILLIAM SOUND AND THE GULF OF ALASKA

B. Study ID Number:

C. Project Leaders: Harold Kibby, Project Officer
USEPA Environmental Research Lab
Corvallis, OR 97333, FTS 420-4625

Richard Meganck, Co-Principal Investigator
Man Tech Environmental
USEPA Environmental Research Lab
Corvallis, OR 97333, FTS 420-4805

Jay D. McKendrick
Co-Principal Investigator/Senior Scientist
Department of Agronomy, University of Alaska
Fairbanks, Alaska 99775, TEL (907) 745-3257

D. Lead Agency: USEPA Environmental Research Lab
Corvallis, OR 97333, FTS 420-4600

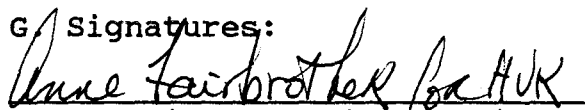
Collaborating Agency: University of Alaska Fairbanks, School
of Agriculture and Land Resources Mgmt.

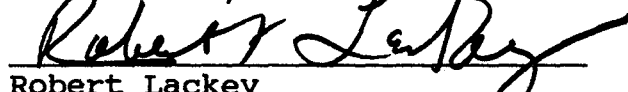
E. Cost of Proposal: FY91 \$133,973
FY92 \$100,000
FY93 \$50,000
FY94 \$50,000
FY95 \$50,000

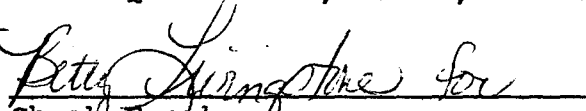
TOTAL COST OF PROJECT: \$383,973

F. Inclusive Dates of Study Plan: FY91-95

G. Signatures:


Harold Kibby, Project Officer
USEPA ERL-C, FTS 420-4625


Robert Lackey
USEPA QA Officer, ERL-C, FTS 420-4601


Chuck Frank
USEPA Financial Officer, ERL-C, FTS 420-4651

FEASIBILITY OF RESTORING THE BAY OF ISLES AND TONSINA BAY IN PRINCE WILLIAM SOUND AND THE GULF OF ALASKA

II. INTRODUCTION

In March 1989 the *Exxon Valdez* ran aground on Bligh Reef in Prince William Sound, spilling approximately 11 million gallons of Prudhoe Bay Crude Oil into the water and impacting over one thousand miles of coastal resources in the Prince William Sound (PWS) and the Gulf of Alaska (GOA). The *Exxon Valdez* oil spill (EVOS) affected the region's environmental habitat, including both floral and faunal populations, as well as recreational, educational, and aesthetic attributes.

Tidal marshes have been classified as the most sensitive shore-type to oil pollution (Ganning et al., 1984). It has been estimated that 2-20 years are required for tidal marshes to recover naturally (Cairns and Buikema, 1984; RPWG, 1990). Oil is rapidly buried in marshes because they are low energy systems, and degradation is limited under the anaerobic conditions found in these environments (Cairns and Buikema, 1984).

Natural marsh recovery begins when oil toxicity is reduced to a point that can be tolerated by recolonizers (Baker et al., 1990). Full tidal marsh recovery hinges on reduction in oil toxicity; availability of propagules; stability of sediments; and biotic interactions (Getter et al., 1984). Restoration activities in heavily oiled marshes may be expected to require both substantial effort and extended time periods. The presence of oil in high concentrations at a site may complicate restoration efforts, and vegetation regrowth in these areas may occur slowly, if at all.

The coastal areas in the PWS and the GOA consist of varied rocky shores, with many small inlets and coves. Tidal marshes represent a relatively small percentage of the coastline affected by EVOS, however they are important components of the coastal ecosystem. Some of these wetlands occur in groundwater discharge areas where the constant supply of water supports high organic soil content and water-loving plants (for example *Carex* spp). It is likely that natural groundwater discharge will cleanse the sediments over time, but increasing the groundwater flow rate may accelerate the cleansing process which, in turn, may assist the rate of recovery for restored areas. If a simple procedure can be shown to effectively increase groundwater discharge which in turn increases the rate of contaminant removal, and if this improved rate of cleansing is confirmed by an accelerated rate of revegetation, the technique may have application in other impacted areas.

A restoration planning process was initiated in late 1989 to begin addressing ways to help restore resources impacted by the

EVOS. A restoration planning progress report, prepared by the interagency Restoration Planning Work Group (RPWG) was released in August 1990. This report defines restoration as "actions undertaken to return an injured resource to its baseline condition, as measured in terms of the injured resource's physical, chemical, or biological properties or the services it previously provided." (RPWG, 1990). Further, Title VII, Oil Pollution Research and Development Program, of the Oil Pollution Act of 1990, (P.L. 101-380) specifically notes the importance of "research and development of methods to restore and rehabilitate natural resources damaged by oil discharges ... and the preparation of scientific monitoring and evaluation plans" for these areas.

There are two tidal marshes, the Bay of Isles and Tonsina Bay, that are still heavily oiled and lacking in natural regeneration of oil-affected vegetation. (see attached maps). A qualitative survey conducted over 4 days in August of 1990 noted (1) heavy effects from residual oil (approximately one acre of mixed Carex and Triglochin) and suspected effects to 1/4 to 1/2 acre of Zostera at the Bay of Isles, and (2) extensive effects to Puccinellia from residual oil at Tonsina Bay; Glaux and other species at higher elevations in this marsh were not apparently affected. See Attachment A for maps of Prince William Sound and the Gulf of Alaska, as well as detailed maps of the Bay of Isles and Tonsina Bay showing oiled areas as based on the oil spill maps.

III. OBJECTIVES

The goal of this feasibility study is to determine whether or not transplanted vegetation can be established and/or enhanced at the Bay of Isles and Tonsina Bay restoration sites, two tidal marshes known to be heavily impacted by oil. Inherent within this goal are the following objectives:

- 1) Quantitatively determine the degree of revegetation success (proportional survival/plot) and relate to crude oil degradation patterns using spatial analysis techniques at both sites.
- 2) At the Bay of Isles test site, increase the rate of groundwater discharge to a small section of the wetland in order to determine if sediment characteristics improve more rapidly where the rate of groundwater discharge has been increased than in areas where the rate of groundwater discharge is normal.
- 3) Demonstrate whether revegetation success rates improve in areas where the rate of groundwater discharge is increased as opposed to areas where the rate of groundwater discharge remains normal.

- .4) Monitor overall site revegetation success on an annual basis. On plots where there is no revegetation success, replant at the same densities in the succeeding year(s).

IV. METHODS

Plantings: Site restoration will consist of conducting restoration trials, and monitoring. The first annual planting will occur in the spring of 1991. Stands will be established using species native to Prince William Sound and the Gulf of Alaska. Each Bay will have twenty-four 10 m² rectangular plots stratified by major substrate type. These will be delineated, marked with aluminum-capped rebar and revegetated with either bare-root or plug transplantings to a uniform density (nine plantings/m²). Broome et al. (1986) discovered that Spartina alterniflora planted 45-60 cm apart were more successful than if spaced farther apart.

Groundwater flushing: In terms of the hydrological component of the project to be undertaken at the Bay of Isles test site, the rate at which water will infiltrate the soil just above the wetland will be determined. This can be done by using falling-head or constant-head parameters (Novitzki, 1976). Next, estimate the rate of flow in the nearby stream at the time of the field visit, and compare that to a nearby long-term streamflow record in order to estimate the approximate low flow expected in the stream during the period of the study (Novitzki, 1979). Use these data to determine the amount of water available from the stream for creating recharge and the size of impound area necessary to allow the water to infiltrate. Use pipe or flexible hose to intercept water from a nearby stream, at an elevation five to ten feet above the mean high water shore line of the wetland, and transport the water to a small, shallow impoundment at the upper edge of the wetland.

The stream end of the pipe will be anchored at a protected location in a pool, preferably just upstream of a rock riffle. The intake point should be five to ten feet above the elevation of the edge of the wetland to allow water to flow by gravity to the recharge site. The inlet will be protected by a screen or grate to allow a reasonable intake of water for extended periods without maintenance. The pipe or hose will be anchored along its length. No effort will be made to protect the system from freezing because increasing recharge (and consequently discharge) during the warm months will be adequate to demonstrate the effectiveness of this technique.

The recharge area will be created by shoveling loose gravel to make a berm 12 to 18 inches high. The soil will be shoveled from the uphill side so that the gravel removal area and berm together form a shallow basin. The basin should be at least 25 but no more than 75 feet long, and from 5 to 10 feet wide. The location of the

outlet end of the pipe will be moved up and down hill (at the edge of the recharge area) until the flow rate approximates the desired recharge rate. The flow will be measured volumetrically, using a calibrated container and stop watch.

Approximately 15 days work will be required at the Bay of Isles¹ and Tonsina Bay for initial site delineation, characterization, installation of the hydrological equipment, and planting in the spring of 1991, with 5 days of follow-up monitoring in the early fall of 1991. This estimate is based on 5 people (eight to ten hour workdays) for the initial planting and 2 people for the monitoring phases. If all plots exhibit plant survival, future activities will be limited to monitoring restoration success and will require 5 days of field work in the early spring and 5 days in the fall for approximately 4 years following installation. Plots which exhibit little revegetation success will be replanted during the 1992 field season. Additional time will be required to collect material and replant any plots on which no plants survived. Care will be taken not to injure sites with equipment or foot traffic. Restoration activities conducted under this project will not interfere with ongoing projects in Prince William Sound and the Gulf of Alaska.

IVa. Identification of Donor Site

Field observations in the summer of 1990 have identified several potential donor sites (transplant sources for revegetation) for the Bay of Isles and Tonsina Bay:

- 1) Tidal marsh at the head of Outside Bay on Naked Island, (Carex)
- 2) Tidal marsh on Crafton Island, (Carex)
- 3) East Bay tidal marsh on Perry Island, (Carex)
- 4) Tonsina Bay, (Puccinellia)
- 6) Fringe tidal marsh around the Bay of Isles and Marsh Bay on Knight Island.

Although these sites may have potential as donor sites, they have not been investigated in detail. Therefore, the following information will be collected to evaluate potential donor sites:

- Species present - The composition of a tidal marsh will factor into its potential to serve as a donor site, based on the species requiring replacement at the Bay of Isles and Tonsina Bay. The site must also have an abundant supply of the appropriate species for revegetation of the Bay of Isles and Tonsina Bay.

¹ Dependent on obtaining permission from adjacent land owner.

- Oil impact - A donor site must be an "unstressed" system (void of unnatural perturbations outside of natural stress), and therefore lacking in any apparent impact from oil.
- Historical treatment record - Again, since a donor site must be "unstressed" relative to the Bay of Isles and Tonsina Bay, a potential donor must not have been subjected to any type of treatment or cleanup operations.
- Vigor - To qualify as a donor site, a tidal marsh must exhibit nearly 100% cover of healthy vegetation, again demonstrating the importance of an "unstressed" system.
- Proximity of vegetative donor site - It is cost-effective and ecologically prudent to choose a donor site in close proximity to the Bay of Isles and Tonsina Bay.
- Size of donor plot - The donor site must be large enough that collection of plants for transplantation will not adversely affect the donor. Less than 1 percent cover will be removed from each donor site.

IVb. Revegetation

Plants will be installed within 48 hours after being collected. The revegetation alternatives selected are bare root, and plugs. Revegetation techniques for Carex involve collecting bare root plants from donor sites, bundling them in groups of 3, and replanting as soon after collection as is feasible. For Puccinellia, it has been shown that plugs survive and grow better than sprigs, so plugs will be used for site restoration (Seneca et al., 1982). When using Puccinellia transplants for restoration, it is important to sufficiently drain the plants.

All transplanted material will be fertilized at the time of installation. According to Broome (1989) transplants usually benefit from fertilizer the first growing season. Either slow release or conventional water soluble fertilizers can be used. The most widely used fertilization method is approximately 15-30g per plant of slow release fertilizer (14-14-14 analysis with a 3 month longevity) in the planting hole (Broome, 1989).

Fertilization was shown to greatly increase growth in Zostera marina in a study by Orth (1977). Fertilizer was massaged by hand into the sediment at the beginning of the experiment (repeated twice) and resulted in a large increase in leaf growth. Studies of tidal marshes affected by the Amoco Cadiz oil spill by Seneca et al. (1982) indicated fertilization (using Mag-Amp and Osmocote) was necessary for significant plant growth because cleanup operations had left large areas void of vegetative cover. Seneca

et al. (1982) also observed higher cover for fertilized Puccinellia transplants.

IVc. Site Restoration Activities

The following activities will be conducted for site restoration at the Bay of Isles and Tonsina Bay:

- (1) Provide a site description by completing a Tidal Marsh Restoration Data Form (see attachment A), including notations of the amount of oil originally estimated to be present in the area, according to the oil spill maps (i.e., light, moderate, heavy).
- (2) Set up twenty-four 10 m² rectangular plots at each marsh representing substrate and selected positions of the tidal zones, and establish a reference point. Note all locations and dimensions on the data sheet.
- (3) List the species to be replaced, and calculate the total number of required transplants. Calculate the amount of fertilizer needed based on the total number of transplants required.
- (4) Establish a permanent reference point within each treatment plot and take soil samples, including replicates (number to be statistically determined), using a random number table to choose the sample points. Use a 6.5 cm diameter piston corer, place the sediment into solvent rinsed foil, wrap, and store (Burns and Teal, 1979). Place labels on each sample and code with a unique I.D. number. Place tape over label to ensure it adheres to the sample and does not smear. Place samples in a cooler and transport to lab for analyses. Rinse all utensils with redistilled solvents before reuse (Burns and Teal, 1979).
- (5) The soil samples should be analyzed for organic content, nutrients (total plant-available N,P,K,Ca and Mg), pH, and salinity at the Soil Science Lab at Oregon State University. Total hydrocarbon and weathered hydrocarbon fractions will be analyzed by SAIC Inc. in San Diego, CA. It will be important to relate revegetation success (survival rate) to particular oil fractions present. Tidal marsh species are elevation specific, and this factor may play an important role in establishing a stand for a particular species.
- (6) Photograph the plot from a reconstructible point and log the film frame and roll number.

- (7) Determine an appropriate donor site for both the Bay of Isles and Tonsina Bay and record locations.
- (8) For transplantation of upper tidal marsh vegetation, the following methods will be employed for bare root transplants (based on Spartina alterniflora using Broome, 1989):
 - i) Obtain bare root transplants (Carex) from the edges of the selected donor tidal marshes. Loosen the plants with a shovel and remove from the marsh. Carefully remove sediment from the roots and bundle in groups of three. Using a plastic bag, place transplants in the bag so the roots are covered, and keep the roots moist.
 - ii) To hand plant, work in pairs. The first worker creates a hole with a dibble approximately 15 cm deep, and adds 0.21 lb of fertilizer for one bundle per hole. Fertilizer applications will be pre-measured and bagged in plastic bags at the laboratory. A second worker inserts plants and firms the soil around the plants. For this project, whether there is a need to work in pairs or individually should be determined in the field.
- (9) For transplantation of upper tidal marsh vegetation the following methods will be employed for plug transplants:
 - i) Obtain plug transplants (Puccinellia) from a donor site by inserting a coring device approximately 20 cm into the substrate, and removing the intact plug from the ground.
 - ii) Remove plug from the coring device and place in plastic bags to keep the plug moist during transport.
 - iii) To hand plant, create a hole with a dibble or coring device large enough to hold the plug, insert fertilizer into the hole, and insert the plug. Firm the soil around the plug to anchor it.
- (10) Take a second photograph of the site once the transplants have been planted, and log the film frame and roll number.
- (11) Observations involving biomass, percent cover, and vigor will rely on the experience and professional judgement of the investigator.

IVd. Site Monitoring

The Bay of Isles and Tonsina Bay will require monitoring on a bi-annual basis, at the end of the growing season in the fall of 1991, and the spring and fall of subsequent years. Monitoring

results should be recorded on the Example Tidal Marsh Site Monitoring Data Form (see attachment B).

The following activities will be performed during site monitoring:

- (1) Each site will be visually assessed for survival of vegetative cover within the revegetated plots. Special attention will also be paid to the apparent vigor of the plants. Vegetation may decline in the second and third year after planting, indicating the need for long-term monitoring before successful restoration can be achieved.
- (2) Other measurements will include soil samples from the revegetated plots established at the Bay of Isles and Tonsina Bay, using the same methods in site restoration. The soil samples will again be analyzed for petroleum hydrocarbons, nutrients, pH, organic content, and grain size.
- (3) Use the quadrant method for eelgrass. Collect information on density, number of shoots, and area covered using 0.1 m² quadrants (Simenstad et al., 1989).
- (4) When cover is evenly distributed, sample using three to five 0.5 m² quadrants within the 10 m² circular plots.
- (5) Take pictures of the site to compare with previous site conditions.
- (6) If no vegetation survives until the following monitoring period take soil samples, replant and monitor the site on an bi-annual basis.

IVe. Quality Assurance/Quality Control (QA/QC)

See attached QA/QC document.

IVf. Logistics

Logistics will be difficult in Alaska due to the remoteness of the location, and will include the following:

- (1) One 5 person team will revegetate the sites.
- (2) Fertilizer and all sampling gear will be taken with each trip either by seaplane or boat.
- (3) After nutrient sample results are known, site restoration will begin on those sites. The restoration team will use a small boat or seaplane to travel to donor sites to obtain transplants.

- (4) Similar activities will take place for site monitoring and any supplemental restoration activities.

IVg. DURATION

The duration of the true feasibility study is one year. If revegetation is unsuccessful, however, replanting may need to occur during subsequent years. The site(s) should be monitored annually until they can be successfully revegetated, and then monitored at least 4 years after successful transplantation before restoration "success" should be evaluated. The study is proposed for the 1991 - 1995 period.

V. DATA ANALYSIS

Va. VEGETATION SUCCESS

The scope of the feasibility study encompasses two tidal marshes, the Bay of Isles and Tonsina Bay in Prince William Sound and the Gulf of Alaska. The study involves site restoration, analyses of soil samples, and visual observations of vegetative cover. Structural parameters will be evaluated each season, and qualitative functional parameters will be evaluated at the end of each growing season. The study will not involve full ecosystem structure and function analyses.

Vb. HYDROLOGIC DATA ANALYSIS

Flow from the pipe will be measured several times during the spring field exercise, and once each subsequent visit. The size of the recharge area will be calculated from measurements made after the flow first begins and the pond size stabilizes, near the end of the spring field period, and once each subsequent visit. These two measurements will provide a calculation of the infiltration rate of the soil under prolonged recharge conditions which can be compared to infiltration rates calculated by the falling-head parameters.

Other measurements will be those sediment characteristics which were planned to be measured at revegetation sites. If sediment characteristics improve most, and revegetation is most successful, near the recharge site, and improvement diminishes proportionally away from that site, there will be a clear demonstration that increased groundwater discharge has accelerated sediment cleansing.

VI. SCHEDULES AND PLANNING

A planning meeting will be convened by the project officer with the Co-Principal Investigators and cooperating scientists

immediately following approval of the proposal to prepare detailed plans for both logistical support and field schedules. Logistical support, including purchase and organization of equipment and materials required for field work, scheduling for air flights and boats will be handled in Alaska. The first site visit is tentatively planned for mid May, 1991 in order to assess potential vegetation donor sites as well as to design, delimit and map planting plots at both Tonsina Bay and the Bay of Isles. Planting is programmed for early June and a follow-up monitoring visit is targeted for early September. In addition, one meeting will be held in Corvallis and one in Alaska to review and assess first year data for second year contingency planning. In addition, preliminary results will be reported to the Oil Spill Restoration Planning Work Group, USEPA ERL-C and Region X.

In subsequent years, two field visits are planned, in the early spring and fall periods for purposes of monitoring success. Intermediate and final results will be reported on an annual basis. In addition to these reports, journal articles and papers for presentation at symposia will be produced.

VII. BUDGET

1. UNIVERSITY OF ALASKA FAIRBANKS (Palmer Research Center
Cooperator)

Personnel	Months	\$
Co-Principal Investigator	1.0	
Research Associate (Surveyor)	2.5	
Research Associate (Biologist)	1.0	
Research Associate (Biologist)	<u>1.0</u>	
	5.5	
Salary and Benefit Total		\$26,764
Travel, Supply, Services		<u>\$10,000</u>
		\$36,764
UAF Overhead @ 43%		\$15,809
SUB TOTAL		<u>\$52,573</u>

2. USEPA ERL - CORVALLIS

Personnel	Months	\$
Co-PI	2.0	
Hydrologist	1.0	
Terrestrial Ecologist	1.0	
Salary and Overhead		\$24,000
Travel (including Co-PI)		<u>\$14,000</u>
SUB TOTAL		\$38,000

3. FIELD LOGISTICAL SUPPORT

Charter Aircraft and Boat \$25,000

4. LABORATORY SERVICES

Chemical Analyses \$10,000

5. SUPPLIES AND EQUIPMENT

Fertilizer, Tools, etc. \$ 7,000
OH supplies and equipment* \$ 1,400
SUB TOTAL \$ 8,400

TOTAL (for 1991)**	\$133,973
TOTAL (1992 -95)	<u>\$250,000</u>
GRAND TOTAL (1991-95)	\$383,973

* Examples of relevant field equipment are as follows:

Field data sheets, clipboards, pencils, spades, measuring tapes, camera, thermometers, salinity meters, 6.5 cm diameter piston corers, solvent rinsed foil, cooler packs, coolers, ziploc bags for soil samples, tape, labels, gloves, raingear, log book.

** Funding levels for 1992 and subsequent years are contingent on restoration/monitoring need as determined through assessment of success of the 1991 program. Funding for 1992 is anticipated at the \$100,000 level, and for 1993-95 at approximately \$50,000 each year.

VIII. PERSONNEL QUALIFICATIONS

The team will include 8 persons:

- Project Officer, USEPA ERL-C, Harold Kibby
- Co-PI Natural Resource Specialist, MAN TECH, Richard Meganck
- Co-PI Agronomist, U. of AK., Jay McKendrick
- Research Associate Surveyor, U. of AK, Warren Fiscus
- Research Associate Biologist, U. of AK, Peter Scorup
- Research Associate Biologist, U. of AK, Gwendo-Lyn Turner
- Hydrologist, MAN TECH, Richard Novitzki
- Terrestrial Ecologist, James G. Wyant

See attached resumes for professional qualifications.

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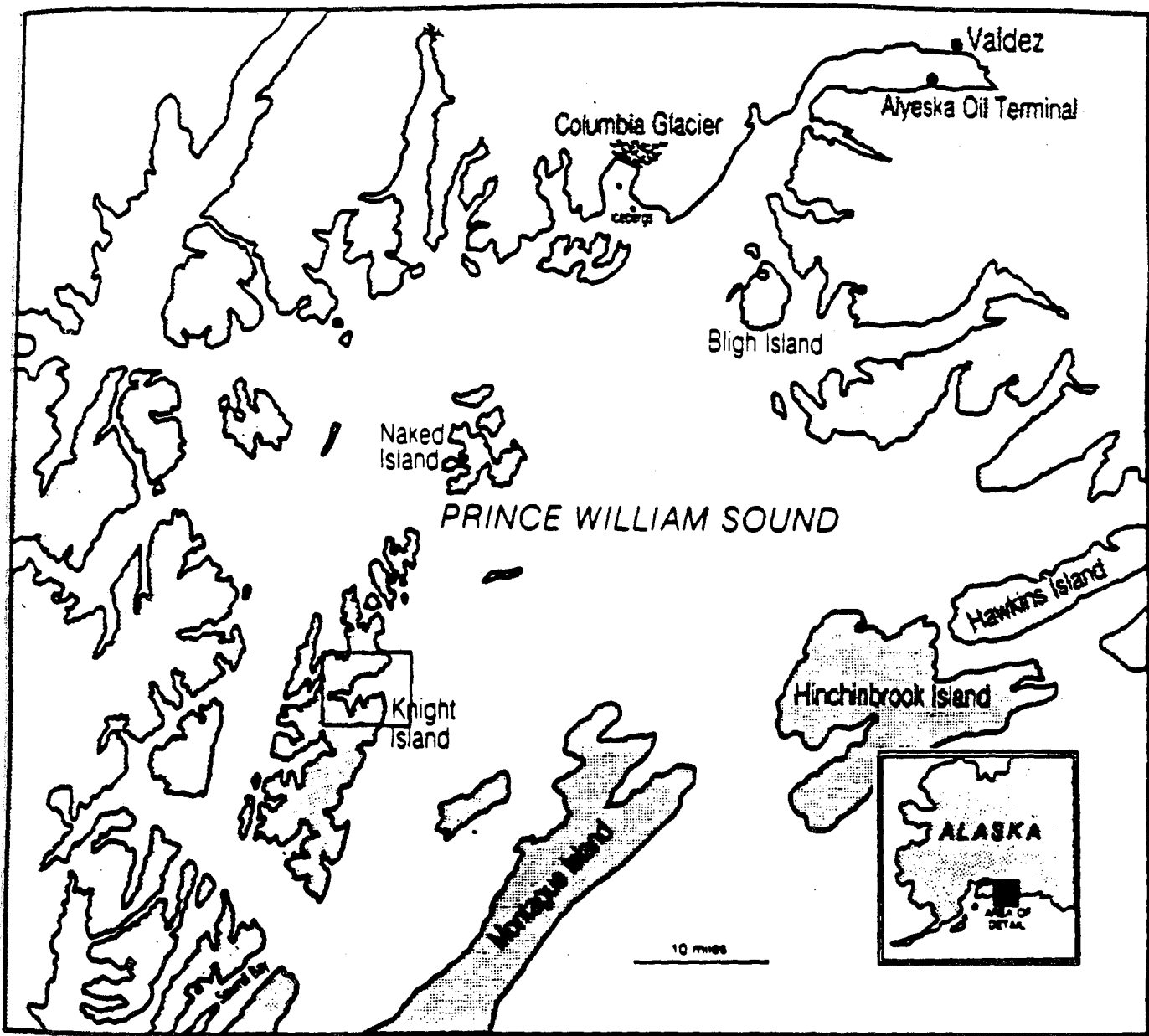
Maps

Prince William Sound

Bay of Isles, Knight Island

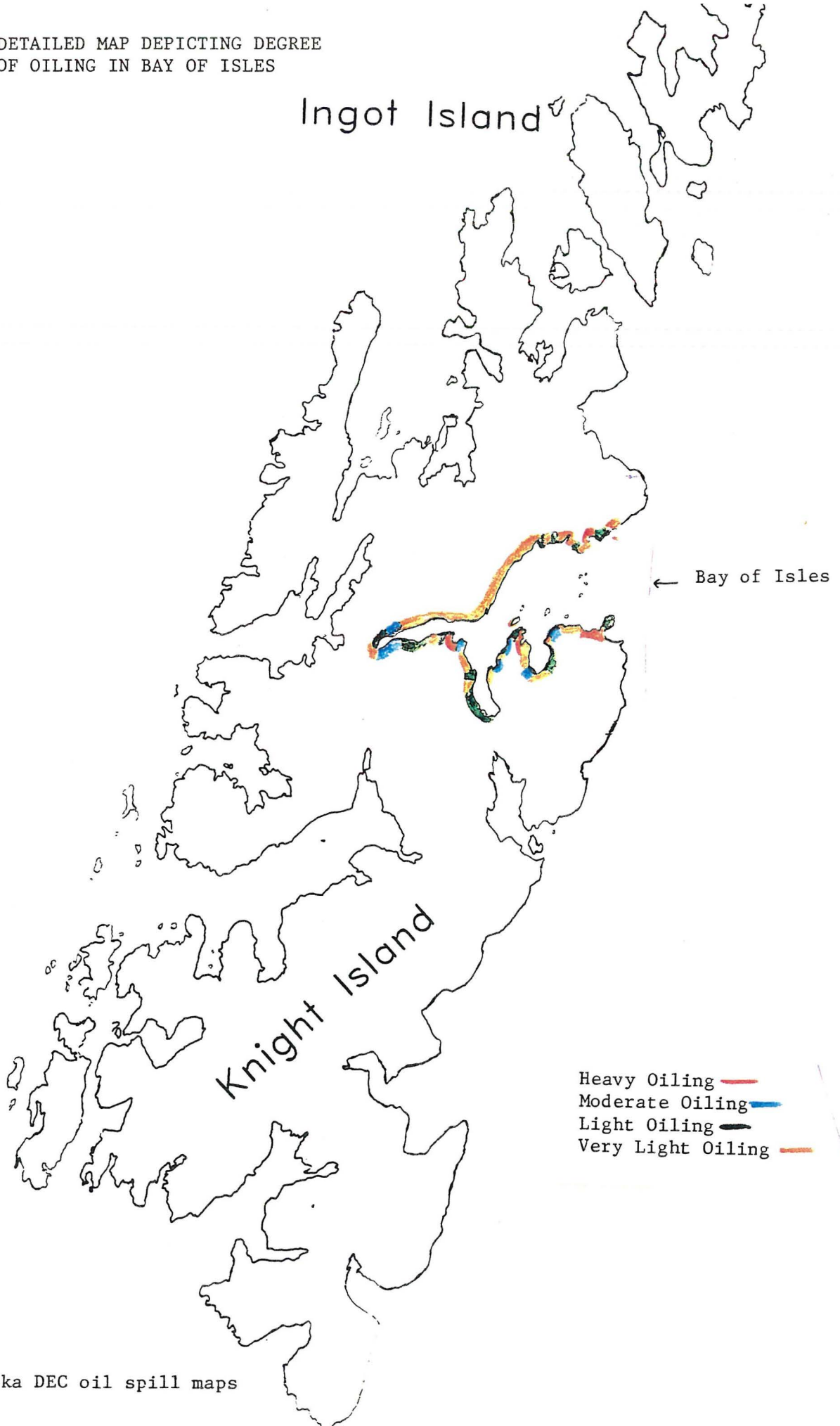
Kenai Peninsula

Tonsina Bay



PRINCE WILLIAM SOUND -- FOR DETAIL OF BLOCKED AREA SEE
DETAIL MAP OF BAY OF ISLES

DETAILED MAP DEPICTING DEGREE
OF OILING IN BAY OF ISLES



SOURCE: Alaska DEC oil spill maps

162°

156°

150°

Bethel

Anchorage

Kenai

Dillingham

ALASKA

Kodiak

Cold Bay

58°

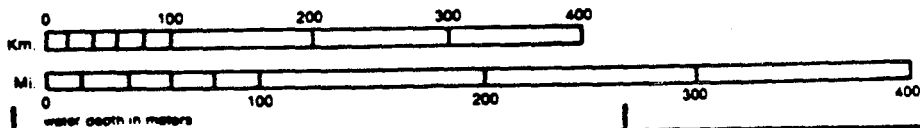
56°

50°

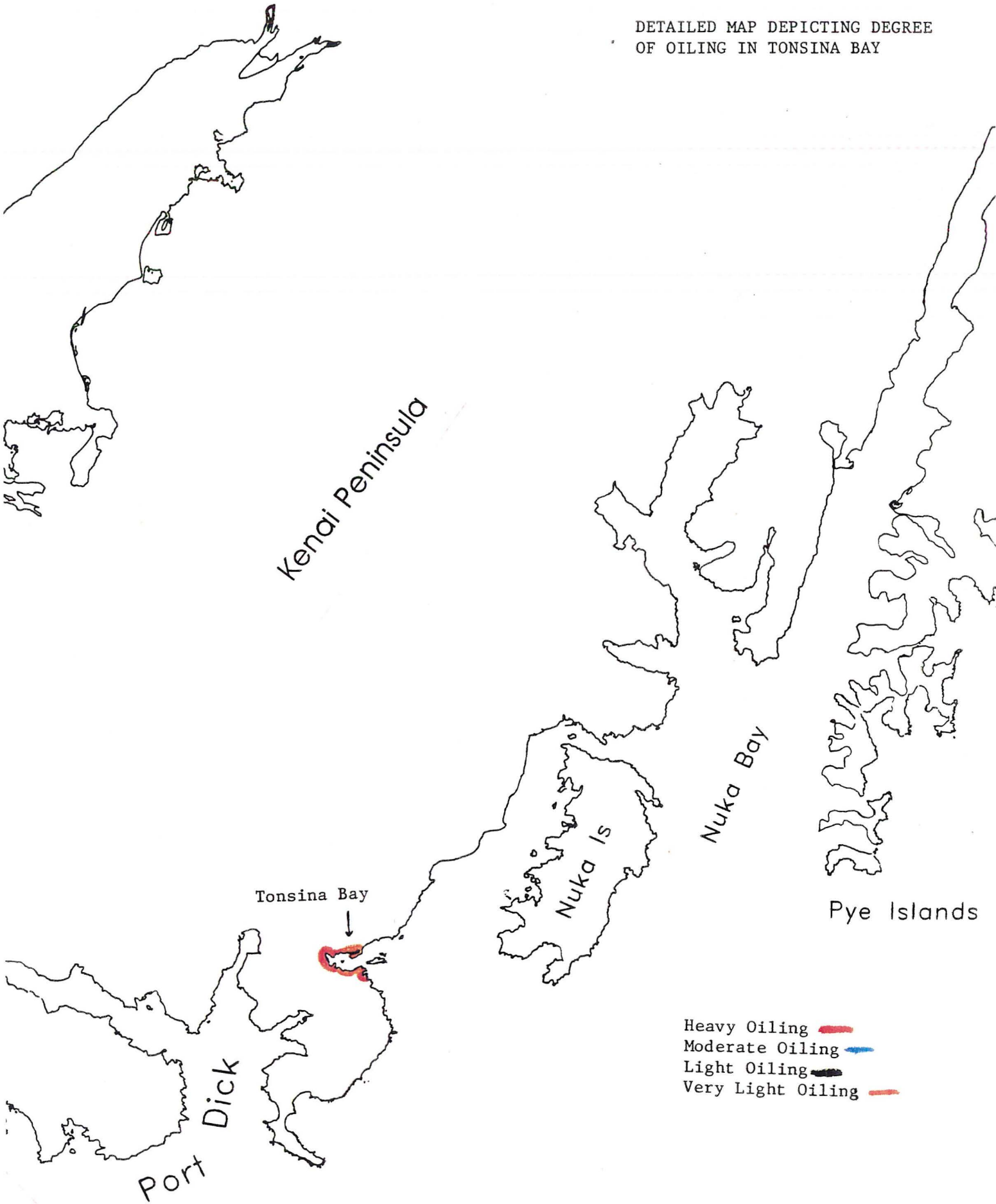
50



KENAI PENINSULA -- FOR DETAIL OF INDICATED AREA SEE DETAIL MAP OF TONSINA BAY



DETAILED MAP DEPICTING DEGREE
OF OILING IN TONSINA BAY



SOURCE: Alaska DEC oil spill maps

ATTACHMENT A

RESTORTION DATA FORM

EXAMPLE TIDAL MARSH RESTORATION DATA FORM

Investigators Name _____
 Date _____
 Time _____
 Location _____
 Segment Number/ID _____

Restoration Assessment

1 Site Description: _____

2 Extent of original oiling (Based on oil map)
 _____ moderate Areal percent _____
 _____ heavy

3 Extent of living/dead vegetation:
 i) Apparent Cover: _____%
 ii) _____% aboveground biomass
 iii) Belowground biomass present beyond limit of aboveground biomass?
 _____ Y _____ N
 if yes, extent of total marsh is _____%;
 location _____

4 i) Location/tidal zone of each treatment area
 Permanent reference point location (landmark) _____
 Distance from reference point (yards) Elevation

#1T _____	#1T _____
#2T _____	#2T _____
#3T _____	#3T _____
#4T _____	#4T _____
#5T _____	#5T _____
#6T _____	#6T _____

5 Approximate area (m²) to be restored per species:
 List: species area

ii) substrate type (S=Sand, SH=Shale, R=Rock/Cobble, M=Mud)
 #1T _____ #4T _____
 #2T _____ #5T _____
 #3T _____ #6T _____

6 Number of transplants needed:
 9 holes/m² @ 3 plants/hole

species	number
i) _____	_____
ii) _____	_____
iii) _____	_____
iv) TOTAL	_____

iii) Location/tidal zone of each control area
 Distance from reference point (yards) Elevation

#1C _____	#1C _____
#2C _____	#2C _____
#3C _____	#3C _____

7 Amount of fertilizer needed: _____ lbs.
 # of plants (6 iv) x .066 lbs. = lbs. of fertilizer needed

iv) Salinity _____‰

8 Vegetative Donor Site:
 i) Proximity to restoration site (approximate miles) _____
 ii) Size (m²) _____
 iii) Donor site identification number _____

9 Comments: _____

10 Soil Analyses (record shipping information on reverse side):
 Soil sample taken? _____ Y _____ N
 i) If yes, number of samples (including duplicates) _____
 ii) I.D. numbers _____
 iii) Method of storage _____

11 Oil characteristics at the site:
 i) surface _____
 ii) subsurface _____
 iii) asphalt _____
 iv) sheen _____

ATTACHMENT B

SITE MONITORING FORM

EXAMPLE TIDAL MARSH SITE MONITORING DATA FORM

Investigators Name _____
 Date _____
 Time _____
 Location _____
 Segment Number/ID _____

<p>1 Restoration method used: i) _____ fertilization ii) _____ transplant/fertilize iii) Date treated _____</p>	<p>2 Species used for each treatment plot: List: #1T _____ #4T _____ #2T _____ #5T _____ #3T _____ #6T _____</p>																																																												
<p>3 Living/dead vegetation cover per treated and control areas: #1T _____ % #2T _____ % #1C _____ % #3T _____ % #2C _____ % #4T _____ % #3C _____ % #5T _____ % #6T _____ %</p>	<p>4 Substrate samples collected for oil/nutrient analysis (Y or N)</p> <table style="width:100%; border-collapse: collapse;"> <thead> <tr> <th></th> <th style="text-align: center;">Oil</th> <th style="text-align: center;">Nutrient</th> </tr> </thead> <tbody> <tr><td>#1T</td><td>_____</td><td>_____</td></tr> <tr><td>#2T</td><td>_____</td><td>_____</td></tr> <tr><td>#3T</td><td>_____</td><td>_____</td></tr> <tr><td>#4T</td><td>_____</td><td>_____</td></tr> <tr><td>#5T</td><td>_____</td><td>_____</td></tr> <tr><td>#6T</td><td>_____</td><td>_____</td></tr> </tbody> </table>		Oil	Nutrient	#1T	_____	_____	#2T	_____	_____	#3T	_____	_____	#4T	_____	_____	#5T	_____	_____	#6T	_____	_____																																							
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<p>5 Apparent vigor</p> <table style="width:100%; border-collapse: collapse;"> <thead> <tr> <th></th> <th style="text-align: center;">1T</th> <th style="text-align: center;">2T</th> <th style="text-align: center;">3T</th> <th style="text-align: center;">4T</th> <th style="text-align: center;">5T</th> <th style="text-align: center;">6T</th> <th style="text-align: center;">1C</th> <th style="text-align: center;">2C</th> <th style="text-align: center;">3C</th> </tr> </thead> <tbody> <tr> <td>i) Vigorous¹ (%)</td> <td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td> </tr> <tr> <td>ii) Healthy² (%)</td> <td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td> </tr> <tr> <td>iii) Low³ (%)</td> <td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td> </tr> <tr> <td>iv) Poor⁴ (%)</td> <td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td> </tr> <tr> <td>v) Dying⁵ (%)</td> <td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td> </tr> </tbody> </table>			1T	2T	3T	4T	5T	6T	1C	2C	3C	i) Vigorous ¹ (%)	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	ii) Healthy ² (%)	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	iii) Low ³ (%)	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	iv) Poor ⁴ (%)	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	v) Dying ⁵ (%)	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>
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<p>6 Comments:</p>																																																													

- Key: 1 (healthy color, >80% cover)
 2 (healthy color, 20-80% cover)
 3 (healthy color, <20% cover)
 4 (unhealthy color)
 5 (unhealthy color - brown stems; sparse cover)

Resumes of PWS/GOA Restoration Team Members

Richard A. Meganck
Natural Resource Planning and Management Specialist

1963 NW Shattock Pl.
Corvallis, OR 97330
H: (503) 757-7557

EDUCATION:

PhD, Oregon State University, June, 1975
Major Field: Natural Resource Management
Minor Fields: Regional Planning, Land Use Planning
Fulbright Scholar, Fluent in Spanish

MSc, Michigan State University, January, 1971
Resource Development and Policy, Watershed Management

BSc, Michigan State University, January, 1968
Park Administration and Planning

EXPERIENCE:

Private Consultant (April 1989 - present)

Man Tech Environmental Sciences, Senior Research Scientist/
Ecological Restoration Project Manager, U.S. Environmental
Protection Agency Research Lab, Corvallis, OR., July, 1990 -
present. Develop and manage a national/ international
ecological restoration research and development program.
Establish and test restoration standards in a wide range of
ecosystems. Develop private/public funding base and public
awareness program for specific projects, proposal preparation,
administer projects and professional staff.

Inter-American Development Bank (IDB), Senior Specialist,
Environmental Protection Division, Washington, D.C., December,
1989 - June, 1990. Developed a process for classifying all
proposals submitted to the Bank for funding consideration on
the basis of their environmental costs/benefits. Worked with
economists to draw analytical conclusions on the feasibility
of investment projects.

U.S. Agency for International Development (USAID), Natural
Resources Advisor, Latin American and Caribbean Bureau,
Washington, D.C., April - November, 1989. Prepared a series
of technical inputs and reports concerning natural resource
management technical assistance projects/policies.

Organization of American States, Department of Regional
Development (OAS/DRD), September, 1979 - March, 1989.
September, 1979 - December, 1981 Project Chief -Saltillo,
Mexico: Integrated Economic Development Planning of the San
Lorenzo Canyon. Developed watershed planning model with
forestry, soil conservation, rural development and national park

components. Prepared policy and funding proposal; **January, 1982 -October, 1983** Project Chief - Trinidad and Tobago: Establishment and Management of a System of National Parks and Forests. Coordinated interagency team in preparing a regional land-use plan for the Tacarigua River Valley including forestry, national parks, agricultural diversification, and nearshore marine management. Prepared funding proposal; **November, 1983 -July, 1986** Principal Natural Resource Specialist. Provided technical support to 19 natural resource planning projects in the Caribbean and northern South America contributing to their design, budgeting, implementation and evaluation; **August, 1986 - March, 1989** Assistant Division Chief - Caribbean/Latin America. Administrative and technical responsibility for 50 natural resource/economic development projects, 15 professionals, 9 support staff, 400 consultants, annual budget of \$12.5 M.

Assistant Professor, Department of Recreation Resource Management, College of Forestry, Oregon State University, September, 1975 - September, 1979. Taught resource policy, park planning, wilderness management, regional planning. Investigator on two research projects, advising-internship coordinator.

Visiting Professor, Department of Park Administration, University of Wyoming, Summer, 1977. International travelling seminar: Natural and Cultural Areas of Latin America (Costa Rica, Panama, Peru, Paraguay, Argentina, Brazil, Venezuela).

Peace Corps Volunteer, Colombia, S.A., 1971-72. Assigned to INDERENA as a resource planner and policy specialist. Field work in the Sierra Nevada, Isla de Salamanca, Purace and Tayrona National Parks and La Macarena National Forest.

Planner, Michigan Department of Natural Resources, October, 1970 -March, 1971. Evaluate local and country park development plans.

MEMBERSHIPS/SPECIAL QUALIFICATIONS:

American Association of Geographers
National Recreation and Park Association
Society of American Foresters
Society for Ecological Restoration
Soil Conservation Society of America
The Nature Conservancy

Author of more than 35 journal articles and reports.

Appointed Affiliate Faculty Member, University of Idaho, Department of Wildland Recreation Management, October, 1989.

Appointed Courtesy Associate Professor, Department of Geosciences, Oregon State University, October, 1990.

Fulbright Scholar, Colombia, S.A., June, 1974 - March, 1975. PhD thesis research with Colombia's Renewable Natural Resource Development Institute (INDERENA): "Colombia's National Parks: An Analysis of Management Problems and Perceived Values".

Graduate, National Outdoor Leadership School, 1969; Outward Bound, 1973; International Seminar on National Parks, 1976.

More than 85 technical/administrative trips in the Americas.

PUBLICATIONS AND TECHNICAL REPORTS:

Meganck, Richard A. Colombia's National Parks. 1977. Colombia Today. Vol. XII, No.1.

Wetterberg, Gary B. and Richard A. Meganck. Colombia's National Parks and Related Reserves: An Analysis of Research Needs and Management. 1978. In: International Experiences with National Parks and Related Reserves. Department of Geography, University of Waterloo Pub. Series No. 12 pp 175-232.

Meganck, Richard A. Battle for a Colombian Park. Oryx. Nov., 1978. Vol. XIV, No. 4, pp 352-358.

Meganck, Richard A. and J. Martin Goebel. Shifting Cultivation: A Problem for Latin American National Parks. Parks. Jul.-Sep., 1979. Vol. IV, No. 2, pp 4-8.

Meganck, Richard A. and Kenneth C. Gibbs. A Methodology Applied to the Analysis of Selected Grazing Management Strategies and Dispersed Recreation. U.S. Forest Service Consultant Report. Aug., 1979.

Meganck, Richard A. San Lorenzo Canyon: A Slice in Time. Chihuahuan Desert Research Journal. Sep., 1980. No. 8, pp 6-7.

Meganck, Richard A. and Julio Carrera Lopez. Multiple Use Management Plan for the San Lorenzo Canyon, Saltillo, Mexico. Mar., 1981. OAS Final Project Report.

Meganck, Richard A. Planning for Ecodevelopment in the Chihuahuan Desert. Parks. Jan.-Mar., 1981. Vol. V, No. 4, pp 4-8.

Meganck, Richard A. and Janet O. Meganck. Implicaciones de la Utilizacion de los Sensores Remotos en Países en Desarrollo. (Implications for the Utilization of Remotely Sensed Data in Developing Countries). Geografia. Jul., 1981. No. 93.

Goebel, J. Martin and Richard A. Meganck. Mexico's National Parks: An Update and Management Problem Analysis, 1981. In: Woodpower I -New Perspectives on Forest Usage. Pergamon Press, N.Y. pp 137-151.

Meganck, Richard A. Interrelaciones Entre un Asentamiento Humano Creciente y el Ordenamiento Territorial: Un Ejemplo de una Zona Arida Mexicana (Interrelations Between and Expanding Human Settlement and Land Use Planning: An Example from an Arid Zone in Mexico) 1981. In: Gestion Integrada de Asentamientos Humanos en el Marco Regional. October. Monterrey, Mexico, pp 29-37.

Meganck, Richard A. and Richard E. Saunier. Managing Our Resources. 1983. The (Caribbean) Naturalist. Vol.4, No.8, 13pp Port of Spain, Trinidad and Tobago.

Meganck, R., S. Persad and L. Kisto. Action Plan for the Caura Valley, 1983. The (Caribbean) Naturalist. Vol. 4, No. 9, 15pp Port of Spain, Trinidad and Tobago.

Meganck, R., S. Persad and L. Kisto. Management and Development Plan for the Caura Valley. Final Project Report OAS-DRD/Division of Forestry, Trinidad and Tobago, Govt. Printry. Apr., 1983.

Anonymous. The Health of the Tacarigua River, 1983. The (Caribbean) Naturalist. Vol. 4, No. 9, Port of Spain, Trinidad and Tobago.

Meganck, Richard A. Watershed Management Planning in an Arid Environment. 1984. In: Landscape Planning. by William Marsh. Addison Wesley Pub., Redding MA., pp 140-143.

Meganck, Richard A. The San Lorenzo Canyon Study, Mexico, 1984. In: Integrated Regional Development Planning: Guidelines and Case Studies from OAS Experience. Department of Regional Development, Organization of American States, Washington, D.C. pp. 201-219.

Taylor, Jeremy (Editor). Interview with Richard Meganck. "Protection is Part of Development". The Caribbean Chronicle, Feb.-Mar., 1984. Vol. 99, No. 1578.

Meganck, Richard A. and Bal S. Ramdial. Trinidad and Tobago Cultural Parks. Parks. Apr.-Jun., 1984, Vol. IX, No. 1, pp 1-5.

Huber, Richard M. Jr. and Richard A. Meganck. Natural Areas: The Building Blocks of Development. 1985. The (Caribbean) Naturalist. Vol. 6, No. 1, Port of Spain, Trinidad and Tobago.

Meganck, Richard A., Craig MacFarland and Richard M. Huber, Jr. Institutional Analysis and Arrangements for the Establishment and Management of a National Parks and Protected Areas Program in Grenada, 1985. 58pp. OAS-DRD, Washington, D.C.

Meganck, Richard A., Craig MacFarland and Richard M. Huber, Jr. Policy Statement for the Establishment and Management of a System of National Parks and Protected Areas, 1985. 45pp. OAS-DRD.

Meganck, Richard A. and Indra Furlong-Kelly. History of the Savannah, 1986. The (Caribbean) Naturalist. Vol. 6, No. 9, pp 4-11. Port of Spain, Trinidad and Tobago.

Sanderson, H. Reed, Richard A. Meganck and Kenneth C. Gibbs. Range Management and Scenic Beauty as Perceived by Dispersed Recreationists, 1986. Journal of Range Management, Vol. 39, No. 5, pp 464-469.

Saunier, Richard E. and Richard A. Meganck. Compatibility of Development and the In-Situ Maintenance of Biological Diversity in Developing Countries. Commissioned Paper for: Technologies to Maintain Biological Diversity, U.S. Congress, Office of

Technology Assessment, OTA-F-330, March, 1987.

Saunier, Richard E., Richard M. Huber, Jr. and Richard A. Meganck. Current Status and Management Recommendations for the St. John's River Watershed, Grenada, 1987. Final Project Report OAS-DRD Integrated Development Project. 35pp. Washington, D.C.

Rojas, Eduardo and Richard A. Meganck. Land Distribution and Land Development in the Eastern Caribbean, 1987. Journal of Land Use Policy. Butterworth Pub., England. Vol. IV, pp 157-167.

Huber, Richard M. Jr. and Richard A. Meganck. National Parks of Trinidad and Tobago. 1987. The (Caribbean) Naturalist. Vol. 7, No. 3, Port of Spain, Trinidad and Tobago, pp 3-29.

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Freed, Michael D., Richard M. Huber Jr., Richard A. Meganck and William B. Possiel. Environmental Education/Interpretation in Trinidad and Tobago. 1988. National Association of Interpretation, Research Monograph.

Rojas, Eduardo and Richard A. Meganck. Affecting Rural Displacement Through Land Distribution Policies in the Eastern Caribbean. 1988. Urban Law and Policy Journal. Elsevier Science Pub., N. Holland. Vol. 9, pp 53-67.

Meganck, Richard A., George Vincent and Richard Huber. The Management Challenge of Grand Anse Beach, Grenada. 1988. Proceedings of the International Workshop on Impact Assessment for International Development. Barbados, West Indies, pp 280-291.

Green, Kenneth, Arthur Heyman, Richard Huber, Richard Meganck and Thomas Riegert. Feasibility Proposal for the Development of The Pitons National Park. 1989. OAS/DRD. Final Project Report. 101pp.

Meganck, Richard A. and J. Martin Goebel. Building Bridges of Understanding About the Tropics: An Opportunity for Interpreters. 1989. Journal of Interpretation. Vol. 13, No. 4.

Meganck, Richard A. White-Knuckling It Through A Job Search. 1990. Journal of Interpretation. Vol. 14, No.1.

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Meganck, Richard A. Coastal Parks as Development Catalysts: A Caribbean Example. 1990. Ocean and Shoreline Management Journal. London, England. (In Press).

February, 1991

VITAE

NAME: Jay D. Mckendrick

PRESENT POSITION: Professor of Agronomy
University of Alaska Fairbanks
and
Affiliate Faculty Member
University of Idaho College of Forestry,
Wildlife & Range Sciences

ADDRESS: P.O. Box 902
Palmer, Alaska 99645

PHONE: (907) 745-3257 office
(907) 745-3432 home

EDUCATION: B.S., University of Idaho, Moscow, 1963 (Soils)
M.S., University of Idaho, Moscow, 1966 (Range Management)
Ph.D., Kansas State University, Manhattan, 1971 (Agronomy Range Management
and Plant Ecology)

NOMINATIONS TO HONORARY

ORGANIZATIONS: Xi Sigma Pi
Gamma Delta Sigma

PROFESSIONAL

ORGANIZATIONS: Society for Range Management

ALASKA RESEARCH PROJECTS:

Reclamation of Land Damaged by Oil Spills. (1972 - 1974)

This project was industry funded and involved several other investigators with field investigations in southcentral, interior and the North Slope regions of Alaska. Laboratory analyses of oil-affected soils and the mitigation of soil-damaged terrestrial sites as well as documenting recovery of damaged vegetation were project objectives. We were successful in establishing the first revegetation of arctic tundra affected by an oil spill. Two papers were published in Arctic, and several annual reports prepared for industry use.

Tundra Rehabilitation Research. (1972 - 1974)

This was an industry funded project focused on finding plant materials (primarily native Alaskan grasses) and soil treatments for rehabilitating areas disturbed by arctic oil developments. Several others participated in this project. Long-term effects on soil fertility and plant succession are being monitored with support from other sources. All reports were prepared for industry use.

Applying Remote Sensing Technology for Developing Regulations for Off-Road Vehicle Use for a Selected Portion of the Alyeska Pipeline Route. (1975 - 1976)

This project was initiated to assist the BLM in assessing and identifying sensitive areas along the access road of the Trans-Alaska Pipeline route which might be threatened by off-road vehicle and other recreation users after the pipeline was completed. Color infrared air photos in conjunction with field visits were used to prepare 1:250K maps and a technical report.

Compilation of Cold-Climate Oil-Spill Research and Technology. (1975 - 1977)

This EPA sponsored project was to assess the adequacy of available methods and techniques for preventing damage from oil spills to coastal and inland shorelines, and for restoring oil spill damage. The cold-climate focus involved surveying, completed and on-going research in Alaska, Canada, Northern Europe and the USSR. Data were collected from the literature and through personally visiting various researchers and their facilities.

Mineral Nutrient Studies on Arctic Tundra. (1975 - 1978)

This project was part of the NSF sponsored Research on Arctic Tundra Environments (RATE). It included study of natural and artificial fertilization. An M.S. thesis on effects of caribou carrion was completed by John Swanson, University of Idaho. A paper was presented at the AIBS meeting, Athens, Georgia, August, 1978.

Musk Ox Range Evaluation. (1978 - 1980)

This two-year project was sponsored by the National Science Foundation. Objectives were to identify dietary components of musk oxen grazing on the recently established Musk Ox Farm near Unalakleet, Alaska. That information can be used as a basis for determining range condition and trend indicators. Seasonal changes in herbage quality were also monitored. This project commenced in March 1978 and terminated March 1980. A report is in the 1981 issue of *Agroborealis*.

Homer Beef Production Project. (1977 - 1979)

This two-year project was funded by the Alaska legislature. Range, agronomy, and animal science investigators collaborated to define management techniques for reducing winter feeding costs and increasing summer grazing returns from ranching operations on the lower Kenai Peninsula. This project terminated September 30, 1979 and became a part of the Alaska Agricultural Experiment Station's on-going research program. Annual presentations of results were given to Homer area residents. Reports were prepared for *Agroborealis*.

Sand Dune Revegetation Near Northway, Alaska. (1977 - 1979)

This project was sponsored by the Northwest Alaskan Pipeline Corporation and is aimed at determining revegetation technology to stabilize sand dunes exposed during construction in the Tanana River uplands between Tetlin Junction and the Alaskan/Canadian border. All reports were prepared for industry use and not generally distributed.

Natural Succession on Placer Mine Spoils of Interior Alaska. (1979)

Records from past mining activities were used to age various placer mine spoils in the Fairbanks vicinity. Dr. Bonita Neiland and a graduate student, Katherine W. Holmes, were co-investigators on this study. The Office

of Surface Mining USDI funded this project. The student prepared a report for *Agroborealis* and an M.S. thesis.

Bison Range Study. (1979 - 1984)

This is a new project aimed at determining the dietary composition of forage eaten by bison in the Delta Junction, Alaska area. Alaska Department of Fish and Game, U.S. Army, and the U.S. Soil Conservation Service are cooperators. The State of Alaska is the funding source. A report was prepared for the 1982 issue of *Agroborealis*. A paper was presented 15 October 1982 at the Great Plains Wildlife Damage Control Workshop, Lincoln, Nebraska with Dr. Phil Gipson.

Coal Mine Reclamation. (1979 - 1982)

This was a Department of Energy sponsored project that included documenting the natural species which invade coal spoils and to determine which substrate and site characteristics were most beneficial for mine spoil revegetation. Drs. Wm. W. Mitchell, C.L. Ping, and G.A. Mitchell were co-investigators on this study. Also, animal uses on pre- and post-mined sites were determined by Charles Elliott, a graduate student. Annual progress reports were prepared for DOE, and C. Elliott prepared a report for the 1982 issue of *Agroborealis*. Elliott's dissertation was completed May 1984. Several articles are being written from the dissertation and one report appeared in the July 1984 issue of *Agroborealis*.

Susitna Hydroelectric Project. (1980 - 1982)

This was a contract to provide vegetation maps, plant community data and investigations on secondary succession relative to moose habitat for the proposed Susitna Hydroelectric Project in southcentral Alaska. Field work and map-making were done by professionals recruited specifically for that project. Reports were prepared for the contracting company and Dr. Helm prepared a paper for the 1982 issue of *Agroborealis*. Dr. Wm. B. Collins prepared an *Agroborealis* article in 1983. During 1982 two new professionals were added to the project and the entire project was turned over to them. Drs. Helm and Collins gave a paper on succession along the Susitna River at a symposium on vegetation inventorying, July 1984.

Arctophila Revegetation Feasibility Study. (1985 - 1989)

This project is sponsored by Standard Alaska Production Company in cooperation with the U.S. Fish and Wildlife Service. Objectives are to identify life history of Arctophila fulva, an emergent grass in arctic ponds, that is believed to be important for waterfowl. Life history and ecological relationships between the grass, its habitats, and wildlife are being investigated. One graduate student is involved with the life history study. Undergraduate students, visiting scientists and technicians are participating in the field research.

Gravel Pad Vegetation Experiments Arctic Slope Alaska 1989-1999.

This project is sponsored by BP Exploration (Alaska), Inc. Objectives are to determine how to prepare gravel pads to improve the conditions for supporting indigenous plant species and to collect 20 to 40 indigenous plants from xeric sites on the Alaska Arctic Slope into a botanical garden. Seed production and establishment of the plants will be monitored. An abandoned gravel pad has been restructured to accommodate tests of gravel thickness, tillage to loosen packed gravel, additions of silt, and windbreaks to trap snow and provide protection to young seedlings on the elevated gravel surfaces. In addition four abandoned wellsites in the National

Petroleum Reserve in Alaska (NPRA) will be evaluated to compare substrate conditions and plant colonization on those sites following rehabilitation and abandonment. The NPRA sites were first examined in 1984, and plans call for three examinations over the course of this 10-year project.

OTHER PERTINENT ACTIVITIES:

Participated in several industry and government workshops and schools on remote sensing, oil spills, arctic research, and range management policies of federal government and State of Alaska.

Presented oil spill reclamation research at Arctic Pollution Control Training School, Anchorage, Alaska, 13 June 1977 and 5 December 1977. Sponsored by Crowley Environmental Services.

Oil Spill Response Workshop, Anchorage, Alaska, 27-30 November 1977. Sponsored by EPA/NOAA.

Alaska Hydrocarbon Workshop, Woods Hole, Massachusetts, 8-10 April 1978. Sponsored by Department of Energy.

Rangeland Policies for the Future, Tucson, Arizona, 28-31 January 1978. Sponsored by Council on Environmental Quality, USDA & USDI.

Mid course review, Salt Lake City, Utah, 5-7 February 1980. Sponsored by BLM.

Chaired Rangeland Symposium hearings for Alaska State Legislature House of Representative Special Agriculture Committee, Anchorage, Alaska, 4-5 December 1979.

Member of the National Academy of Sciences Committee on Alaskan Coal Mining and Reclamation. That report was submitted to Secretary of Interior during the summer of 1980.

Served as Governor Hammond's Staff Advisory Designee to National Governor's Subcommittee on Range Resource Management, 1979.

Panel I - Coastal Habitats. Oil Spill Restoration Symposium. 26-27 March 1990, Egan Convention Center, Anchorage, Alaska. (presented results of long-term vegetation recovery on oil-damaged coastal tundra vegetation)

Technical Panel - Domestic Policy Council Interagency Task Force on Wetlands, Anchorage, Alaska. 7 September 1990.

Provided consultation on land reclamation and other topics for:

Alaska Pipeline Office, Anchorage, AK
Alaska Department of Commerce and Economic Development, Juneau, AK
Amerada Hess Corp., Anchorage, AK
ARCO Alaska, Inc., Anchorage, AK
Arctec, Inc., Columbia, MD
Argonne National Laboratory, Argonne, IL
Division of Agriculture, DNR, Palmer, AK
Dowl Engineering, Anchorage, AK
Environmental Science & Engineering, Inc. Anchorage, AK
Geophysical Services, Inc., Anchorage, AK
Gulf Interstate Engineering, Houston, TX
Husky Oil NPR Operations, Anchorage, AK

Northern Testing Lab, Inc., Fairbanks, AK
Northwest Alaskan Pipeline Company, Salt Lake City, UT
PEAK Oilfield Service, Co., Anchorage, AK
PSA, Inc., Anchorage, AK
Science Applications, Inc., Boulder, CO
Tryck Nyman and Hayes, Anchorage, AK
U.S. Geological Service, USDI, Anchorage, AK
Yukon Mining Company, Fairbanks, AK

Graduate Student Committees:

John Swanson - M.S. 1979, University of Idaho (Co-advisor)
Katherine W. Holmes - M.S. 1981 UAF, (Co-advisor)
Charles Elliott - Ph.D. 1984, UAF (Advisor)
Thomas Smith - M.S. candidate 1985, UAF
Janice Dobson - Ph.D. 1986-1989, University of Idaho

Teaching Activities:

Introduction to Range Management (ALR 312) fall semester 1986, 1988, 1990
Forest & Range Plant Identification (ALR 393) Spring Semester 1988
Boreal and Tundra Rangelands (ALR 393) Fall Semester 1990 (Class tour to Prudhoe Bay)

AFES Faculty Selection Committees:

Crop physiologist (1972-73)
Range Instructor (1980)
Soil Scientists (1974, 1979, 1982)
Reindeer Range Scientist (1980)
Beef Scientist (1984)
Laboratory Supervisor (1987-1988)
Forage Agronomist (1988) Chairman
Plant Breeder (1988)

AFES committees:

Palmer laboratory building committee (1983)
Publications (1974-1984)
Palmer laboratory advisory (1985)
Range management review (1985)
SALRM promotion and tenure process evaluation (1985)

UAF and other committees:

UA computer advisory committee 1975-1978

USDA Preparation of USDA Alaska Manifesto to suggest research priorities for Reagan administration's consideration (1981).

UAF Faculty Senate 1987-89

Scholarly Activities Committee Faculty Senate (1988-90)

Campus Promotion and Tenure Committee (1990-92)

Ad Hoc Employee Relations Committee (1989-90)

SALRM/CES Merger Committee (1989-90)

Served as a reviewer for:

National Science Foundation (proposals)

Arctic and Alpine Research (journal articles)

Department of Energy (proposals)

Journal of Range Management (journal articles)

Agroborealis (AFES station publications)

Hatch/McIntire-Stennis (proposals)

Alaska Dept. Natural Resources Division of Agriculture (proposals)

Alaska Science and Technology Foundation (proposals)

PUBLICATIONS

Refereed Journals or Books:

McKendrick, J.D. and L.A. Sharp. 1970. Relationship of organic reserves to herbage production in crested wheatgrass. *Journal of Range Management* 23:434-438.

Owensby, Clenton E., Gary M. Paulsen, and Jay Dee McKendrick. 1970. Effect of burning and clipping on big bluestem reserve carbohydrates. *Journal of Range Management* 23:358-362.

McKendrick, J.D. and Loran C. Anderson. 1971. Variation of reserve-starch-granule areas and diameters in Andropogon gerardi rhizomes. *Agronomy Journal* 63:619-620.

Owensby, Clenton E., Jerry R. Rains, and Jay D. McKendrick. 1974. Effects of one year of intensive clipping on big bluestem. *Journal of Range Management* 27(5):341-343.

McKendrick, Jay D., Clenton E. Owensby, and Robert M. Hyde. 1975. Big bluestem and indiagrass vegetative reproduction and annual reserve carbohydrate and nitrogen cycles. *Agro-Ecosystem*. 2(1975):75-93.

Mitchell, William W. and Jay D. McKendrick. 1975. Responses of arctic, boreal and alpine biotypes in reciprocal transplants. IN: Jerry Brown (ed.) *Ecological investigations of the tundra biome in the Prudhoe Bay region, Alaska*. Biological papers of the University of Alaska, Special Report No. 2. p 92-111.

McKendrick, J.D., Valerie Ott, and George A. Mitchell. 1978. Effects of nitrogen and phosphorus fertilization on carbohydrate and nutrient levels in Dupontia fisheri and Arctagrostis latifolia. Chapter 22 IN: Tieszen, L.L. (ed.) *Vegetation and production ecology of an Alaskan arctic tundra*. Springer-Verlag, N.Y. p 509-537.

McKendrick, Jay D, and Wm. W. Mitchell. 1978. Effects of burning crude oil spilled onto six habitat types in Alaska. *Arctic*. 31(3):277-295.

McKendrick, Jay D. and Wm. W. Mitchell. 1978. Fertilizing and seeding oil-damaged tundra to effect vegetation recovery. *Arctic*. 31(3):296-304.

Mitchell, Wm. W., T.E. Loynachan, and J.D. McKendrick. 1979. Effects of tillage and fertilization on persistence of crude oil in a northern soil. *Journal Environmental Quality* 8(4):525-532.

McKendrick, Jay D., George O. Batzli, K.R. Everett, and John C. Swanson. 1980. Some effects of mammalian herbivores and fertilization on tundra soils and vegetation. *Arctic and Alpine Research*. 12(4):565-578.

McKendrick, Jay D, with the Committee on Alaska coal mining and reclamation. 1980. Surface mining in Alaska: an investigation of the surface mining control and Reclamation Act of 1972 in relation to Alaskan conditions. A report prepared by the Committee on Alaskan Coal Mining and Reclamation Board on Mineral and Energy Resources, Commission on Natural Resources, National Research Council, National Academy of Sciences, National Academy Press. Washington, D.C. 328 pp.

Chapin, F. Stuart III, Douglas A. Johnson, and Jay D. McKendrick. 1980. Seasonal movement of nutrients in plants of differing growth form in an Alaskan tundra ecosystem: Implications of herbivory. *Journal Ecology* 68:189-209.

Hanley, Thomas A. and Jay D. McKendrick. 1983. Seasonal changes in chemical composition and nutritive value of native forages in a spruce-hemlock forest, southeastern Alaska. USDA Forest Service Research Paper PNW-#12, Pacific Northwest Forest and Range Experiment Station, Portland, Oregon. 41 p.

Hanley, Thomas A. and Jay D. McKendrick. 1985. Potential nutritional limitations for blacktailed deer in a spruce-hemlock forest, southeastern Alaska. *Journal of Wildlife Management* 49(1):103-114.

Chapin, F. Stuart III, Jay D. McKendrick, and Douglas A. Johnson. 1986. Seasonal changes in carbon fractions in Alaskan tundra plants of differing growth form: implications for herbivory. *Journal of Ecology* 74:707-731.

Herlugson, Christopher J., Jay D. McKendrick and Mary Lou Herlugson. 1985. Selection of garden residues by Alaska moose, Alces Alces, during winter. *Canadian Field-Naturalist* 99(3):389-391.

McKendrick, Jay D. 1986. Final cleanup at selected (1975-1981) wellsites, sampling and testing of waters and bottom muds in the reserve pits and the recording of tundra plant responses on the National Petroleum reserve in Alaska (NPRO), Volume III recording of plant responses. Nueara Reclamation Co., U.S. Geological Survey, Anchorage, Alaska. 225 pp.

Helm, D.J., J.D. McKendrick, and W.B. Collins. 1987 Fertilizer effects on annual grass in wet sedge-grass vegetation site, Susitna Basin, Alaska, U.S.A. *Arctic and Alpine Research* 19(1):29-34.

McKendrick, Jay D. 1987. Plant succession on disturbed sites, North Slope, Alaska, U.S.A. *Arctic and Alpine Research* 19(4):554-565.

Elliott, Charles L., Jay D. McKendrick, and Dot Helm. 1987. Plant biomass, cover, and survival of species used for stripmine reclamation in south-central Alaska, U.S.A. *Arctic and Alpine Research* 19(4):572-577.

Van Horne, Beatrice, Thomas A. Hanley, Rex G. Cates, Jay D. McKendrick. 1988. Influence of serial stage and season on leaf chemistry of southeastern Alaska deer forage. *Canadian Journal of Forest Research*. 18:90-99.

Komarkova, V. and J.D. McKendrick. 1988. Patterns in vascular plant growth forms in arctic communities and environment at Atkasook, Alaska. In: Werger, J.J.A., P.J.M. van der Aart, H.J. During, and J.T.A. Verboeven (eds.) *Plant form and vegetation structure*. SPB Academic Publishing, The Hague, The Netherlands. pp. 45-70.

McKendrick, Jay D. and Katherine W. Holmes. 1989. Plant species on dredge tailings of interior and gavel pads of arctic Alaska. In: Bandopdhay, Sukumar and Frank J. Skudrzyk (eds.). Mining in the Arctic. Proceedings of the 1st International Symposium on Mining in the Arctic, Fairbanks, Alaska, 17-19 July 1989. A.A. Balkema, Rotterdam. pp. 157-165.

Notes:

McKendrick, Jay D. 1987. Arctophila fulva for revegetating arctic wetlands (Alaska). Restoration and Management Notes 5(2):93-94.

University Publications:

McKendrick, J.D. 1966. An investigation of certain vegetation changes over a ten-year period on five Southern Idaho rangeland seedings. M.S. thesis, University of Idaho, Moscow, Idaho. 111 p.

McKendrick, J.D. 1971. Big bluestem and indiangrass vegetative reproduction and critical levels of reserve carbohydrates and nitrogen. Ph.D. Dissertation. Kansas State University, Manhattan, Kansas. 70 p.

McKendrick, J.D., and Peter C. Scorup. 1974. A super bird's-eye view of Alaska. *Agroborealis*. 6(1):26-30.

McKendrick, J.D. 1974. Prospecting for green gold. *Agroborealis*. 6(1):13-14.

Mitchell, Wm. W., J.D. McKendrick, F.J. Wooding, and M.A. Barzee. 1974. Agronomists on the banks of the Sagavanirktok. *Agroborealis*. 6(1):33-35.

Mitchell, George A. and Jay D. McKendrick. 1975. Volcanic-ash-affected soils of southcentral Alaska: some chemical and mineralogical properties. *Agroborealis* 7:21-23.

McKendrick, J.D. 1975. Soil nutrients. In: J. Brown, ed. Ecological and limnological reconnaissances from Prudhoe Bay into the Brooks Range., Alaska. Summer 1975. U.S. Army Cold Regions Research and Engineering Laboratory, Hanover, NH. p 23-23.

McKendrick, Jay D. 1976. Photo-plots reveal arctic secrets. *Agroborealis* 8:25-29.

McKendrick, Jay D., A.L. Brundage, and V.L. Burton. 1977. Quality bluejoint hay is influenced by time of harvest. *Agroborealis* 9(1):26-29.

McKendrick, Jay D., Wm. W. Mitchell, and Fredric M. Husby. 1979. The Homer beef production project - a cooperative effort in applied research. *Agroborealis*. 11(1):4-5.

McKendrick, Jay D. 1979. Hay quality survey for the Homer beef production project - 1977. *Agroborealis*. 11(1):6-10.

McKendrick, Jay D. and David P. Bleicher. 1980. Observations of a grass bug on native bluejoint ranges. *Agroborealis*. 12(1):15-18.

McKendrick, Jay D. 1980. Mine reclamation in portions of West Germany, the Union of Soviet Socialistic Republics and Alaska relative to Alaska. *Agroborealis*. 12(1):11-14.

McKendrick, Jay D. 1981. Responses of arctic tundra to intensive muskox grazing. *Agroborealis* 13:49-55.

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McKendrick, Jay D. 1982. Alaska's bison - A game biologist's range-management problem. *Agroborealis* 14:73-79.

McKendrick, Jay D. 1983. Alaska's rangelands, Chapter 11. IN: Alaska's Agriculture and Forestry, Alaska Rural Development Council Publication #3. pp 125-156.

McKendrick, Jay D. 1983. Potential beef production, Chapter 12. IN: Alaska's Agriculture and Forestry, Alaska Rural Development Council Publication #3.

McKendrick, Jay D. 1983. Alaska's range livestock potential. IN: An economic assessment of Alaskan agriculture. Alaska Department of Commerce and Economic Development, Division of Finance and Economics for the Alaska Agricultural Action Council. Juneau. pp. 138-152.

McKendrick, Jay D., Charles L. Elliott and Charles P. Boddy. 1984. Evaluation of plants used for stripmine reclamation near Healy, Alaska. *Agroborealis* 16(2):5-8.

McKendrick, Jay D. 1984. Range management boreal zone Alaska. University of Alaska Cooperative Extension Service A-00145. Fairbanks. 14 pp.

McKendrick, Jay D. 1985. Animal distribution limits range utilization. *Agroborealis* 17(1)37-40.

Proceedings:

Bonde, E.K., Maxine F. Foreman, T.A. Babb, S. Kjølvik, J.D. McKendrick, W.W. Mitchell, L.L. Tieszen, F.J. Wooding, and W. Younkin. 1973. Growth and development of three agronomic species in pots ("phytometers"). IN: Primary production processes, International Biological Programme, Tundra Biome Proceedings. p. 99-110.

McKendrick, Jay D. 1976. Agrometeorology in northern regions. *Bulletin. American Meteorological Society* 57(1):38-39.

McKendrick, J.D. 1980. Forage Crops. IN: Agricultural opportunity: A Management Prospective, Proceedings. Division of Economic Enterprise, Department of Commerce and Economic Development, State of Alaska, Juneau. pp. 35-38.

Gipson, Philip S. and Jay D. McKendrick. 1981. Bison depredation on grain fields in interior Alaska. Proceedings of The Fifth Great Plains Wildlife Damage Control Workshop, University of Nebraska, Lincoln. (Timm, Robert M. and Ron J. Johnson, eds.) p. 116-121.

Helm, D., W.B. Collins, and J. McKendrick. 1984. Floodplain vegetation succession in southcentral Alaska. IN: Proceedings, Inventorying Forest and other Vegetation of the High Latitude and High Altitude Regions. Fairbanks, Alaska. July 23-26, 1984.

Elliott, Charles L. and Jay D. McKendrick. 1985. Food habits of Dall sheep on revegetated coal stripmine spoils in Alaska. IN: Hoefs, Manfred (editor) Northern Wild Sheep and Goat Council, Proceedings of the Fourth Biennial Symposium. 1984. pp 241-251.

Hanley, Thomas A., Rex G. Cates, Beatrice Van Horne, Jay D. McKendrick. 1985. Forest stand-age-related differences in apparent nutritional quality of forage for deer in southeastern Alaska. *IN*: Provenza, Frederick D.; Jerran T. Flinders and E. Durant McArthur (Compilers). Proceedings--Symposium on plant-herbivore interactions; 1985 August 7-9, Snowbird, Utah. Gen. Tech. Rep. INT-222. Ogden, Utah. U.S. Department of Agriculture, Forest Service, Intermountain Forest and Range Experiment Station; 1987. 179 pp.

Brown, Anne L. and Jay D. McKendrick. 1987. Joint Industry/Agency/University Revegetation Feasibility Project. The Fifth Symposium on Coastal and Ocean Management, May 26-29, 1987, Seattle, Washington.

Komarkova, V. and J.D. McKendrick. 1987. Vascular plant growth forms in arctic communities and environment at Atkasook, Alaska. International Symposium on Vegetational Structure, July 14-18, 1987. University of Utrecht, The Netherlands.

McKendrick, Jay D. 1988. Soil Fertility Observations in Arctic Alaska. 117th Annual Meeting Society of Mining Engineers, 25-28 January 1988, Phoenix, Arizona. pp. 17.

McKendrick, Jay. 1988. Vegetative reproduction of *Arctophila fulva*. 39th Arctic Science Conference Proceedings, American Association for the Advancement of Science -- Arctic Division, 7-10 October 1988, University of Alaska Fairbanks, Fairbanks, Alaska. pp. 200.

Elliott, Charles L. and J.D. McKendrick. 1988. Avian use of reclaimed stripmines in interior Alaska. 1988 National Symposium on Mining, Hydrology, sedimentology, and Reclamation. 5-9 December 1988, Reno, Nevada.

Elliott, Charles L. and Jay D. McKendrick. 1989. Summer food habits of tundra voles (*Microtus oeconomus*) on revegetated coal stripmine spoils in central Alaska. 69th Annual Meeting American Society of Mammalogists. 11-15 June 1989, University of Alaska Fairbanks, Fairbanks, Alaska.

McKendrick, Jay D. 1989. Establishing vegetation on gravel pads in arctic Alaska. First International Symposium on Mining in the Arctic. July 17-19, 1989, University of Alaska Fairbanks, Fairbanks, AK.

McKendrick, Jay D., Kurt C. Nelson, and Christopher J. Herlugson. 1989. Digestibility estimates of winter browse for moose, upper Kenai Peninsula, Alaska. 40th Arctic Science Conference Global Change. September 14-16, 1989, Fairbanks, Alaska. p.43

Dobson, J.L., R. Robberecht, and J.D. McKendrick. 1989. The response of *Arctophila fulva* to rapidly changing aquatic and terrestrial habitats associated with oil development in northern Alaska. Abstract. 74th Annual Ecological Society of America, University of Toronto, Toronto, Ontario. August 6-10, 1989. Bulletin of the Ecological Society of America 70(2):99.

Elliott, Charles L. and Jay D. McKendrick. 1990. Big game use of reclaimed stripmine lands in interior Alaska. 1990 National Symposium on Mining. Knoxville, Tennessee, May 14-18, 1990. University of Kentucky Office of Engineering Continuing Education & Extension. PP 1-5 *IN*: Graves, D.H. (ed), Proceedings National Symposium on Mining, Publication No. UKY BU153, University of Kentucky, Lexington. 275 pp

McKendrick, Jay D. 1990. Seed formation by *Arctophila fulva* in Relation to Temperature, Arctic Coastal Plain, Alaska. June 1990. Symposium on the Role of Arctic Regions on Global Change, University of Alaska Fairbanks, Fairbanks, Alaska. (paper in review for proceedings)

McKendrick, Jay D. 1990. Plenary address. Ecology, Energetics, Bionenergetics, Human Health - ECOBIOEN - 90 International Meeting. Dagomys Hotel, Sochi, USSR. 23-30 September 1990.

Reports:

Institute of Agricultural Sciences (Mitchell, Wm.W. and Jay D. McKendrick), University of Alaska. 1972. Report of Research Progress on Rehabilitation of Disturbed Ground in Arctic Alaska. To: Atlantic Richfield Co., Humble Oil Co., Alyeska Pipeline Service Co., Shell Oil Co., Union Oil Co. 35 p.

Mitchell, Wm.W., J.D. McKendrick, M.A. Barzee, and F.W. Wooding. 1974. Report of research progress on Reclamation of land damaged by oil spills. To: Alyeska Pipeline Service Co. University of Alaska, Institute of Agricultural Sciences, Palmer Research Center. 48 pp.

Mitchell, W.W. and J.D. McKendrick. 1975. Progress Report 1974 Tundra Rehabilitation Research: Prudhoe Bay, and Palmer Research Center. To: Alyeska Pipeline Service Company, Atlantic Richfield Company, Canadian Arctic Gas Study Limited, Exxon Company, Shell Oil Company, Union Oil Company. Institute of Agricultural Sciences, University of Alaska, Palmer Research Center, Palmer, Alaska. 84 p. + Pictorial Appendices.

Mitchell, Wm.W. and J.D. McKendrick. 1975. Reclamation of Land Damaged by Oil Spills. To: Alyeska Pipeline Service Co. Progress Report, University of Alaska, Institute of Agricultural Sciences, Palmer Research Center, Palmer, Alaska. 41 p. + Appendices.

McKendrick, J.D., B.J. Neiland, and K. Holmes. 1980. Natural revegetation of placer mined lands of Interior Alaska II. Contract Report, Mineral Industry Research Laboratory, University of Alaska Fairbanks, Fairbanks, AK. 9p.

McKendrick, J.D., W.B. Collins, and D. Helm. 1980. Susitna Hydroelectric Project Environmental Studies Subtask 7.12 -- Plant Ecology Annual Report. 93 p.

McKendrick, J., W. Collins, D. Helm, J. McMullen, and J. Koranda. 1981. Susitna Hydroelectric Project, Environmental Studies, Subtask 7.12: Plant Ecology Annual Report.

McKendrick, Jay, William Collins, Dot Helm, Joseph McMullen. 1982. Alaska Power Authority Susitna Hydroelectric Project Environmental Studies - Subtask 7.12 Plant Ecological Studies Phase I Final Report. 124 p. + Appendices.

Steigers, William D. Jr., Dot Helm, James G. MacCracken, Jay D. McKendrick, Patrick V. Mayer. 1983. Alaska Power Authority Susitna Hydroelectric Project Environmental Studies - Subtask 7.12 1982 Plant Ecology Studies Final Report. Prepared for LGL Alaska Research Associates, Inc. Anchorage, AK. 288 p + maps.

McKendrick, Jay D. 1985. Progress report outlining activities and significant technical discoveries related to *Arctophila fulva*. University of Alaska Agricultural and Forestry Experiment Station, Palmer, Alaska. 23 pp.

McKendrick, Jay D. 1987. *Arctophila* feasibility study 1986 annual report. Standard Alaska Production Company, Anchorage, Alaska. 69 pp (+ appendices A through H, 352 pp.)

McKendrick, Jay D. 1988. *Arctophila* feasibility study 1987 annual report. Standard Alaska Production Company, Anchorage, Alaska. 90 pp (+ appendices A through H, approximately 350 pp.)

McKendrick, Jay D. 1989. Gravel Vegetation Project 1989 Progress Report. BP Exploration (Alaska), Inc. Anchorage, AK. 4 pp.

McKendrick, Jay D. 1990. *Arctophila* revegetation feasibility study 1988 annual report. BP Exploration (Alaska), Inc., Anchorage Alaska. 59 pp (+ appendices A through K, approximately 180 pp.)

VITAE

Warren Fiscus
P.O. Box 290
Palmer, AK 99645
Phone: 907-745-3257 (work)

SS 523-42-7268
Married - 5 children
High School Graduate

PROFESSIONAL LICENSES: Registered Land Surveyor No. 5572, Colorado (inactive)
Registered Land Surveyor No. 1634-S, Alaska

EXPERIENCE:

1988 - present: RESEARCH TECHNICIAN
University of Alaska Agricultural and Forestry Experiment Station
533 E. Fireweed, Palmer, AK 99645

Responsible for surveying and mapping approximately 115 study sites for the Arctophila Project conducted on the North Slope.

Design, construction layout, and contractor supervision during construction of the Gravel Vegetation Experiment. Other activities include collection of seed, soil, mud, water, plant samples and related data. Designed and built portable seed harvester for use in the collection of native seeds for Gravel Vegetation Experiment.

Prepared soil and plant samples for lab analysis. Threshed, cleaned and ran germination tests on seed collected the past two years.

1972 - 1988: OWNER/OPERATOR of Land Surveying Business

Activities included all incidental duties pertaining to the operation of a business; supervision of employees, financial records, survey records, building and equipment maintenance, drafting, computations, bid and contract writing, subdivision design, cadastral surveys, topography surveys, cross-sections, field surveys, logistics, etc.

1982 - 1985: LHD and Associates
Construction Surveyors
723 W. 6th Avenue, Anchorage, AK 99645

Position - Chief of Parties and Project Supervisor for North Slope Construction Surveying Contract with Arco Oil & Gas Company operations at Prudhoe Bay and Kuparuk oil fields. Duties included supervision of office and field personnel, correlation of survey work with Arco engineers and LHD surveyors during construction of pipelines, module placement, roads and pads. Note this position was a contract with LHD for their contract with Arco Oil & Gas.

1954 - 1972: H.V. Lounsbury and Assoc., 1967 - 1972
State of Alaska, Dept. of Highways, 1965 - 1967
Mid-Continent Coal & Coke Company, 1964 - 1965
Tom Walker, County Surveyor, 1954 - 1964

Positions: Chainman, Instrumentman, Party Chief, Chief of Parties, Inspector, Coal Mine Surveyor, etc.

Passed Land Surveyors examination in Colorado in 1964 for registration and license. Obtained Alaska registration and license in 1967.

GWENDO-LYN TURNER

EDUCATION:

M.S. Ecology. March 1975
University of California, Davis, California.

B.A. Biological Sciences. Minor: Geology. June 1970.
Humboldt State University, Arcata, California.

EXPERIENCE:

7/87 - Present. Research Associate, University of Alaska, Fairbanks, Agricultural and Forestry Experiment Station, Palmer, Alaska. Majority of the work has been with BP Exploration (Alaska) Inc. as field liaison and consultant for North Slope environmental programs. Observing and participating in field sampling programs on the North Slope, reviewing the reports, providing comments and input on results. Programs worked on include: the Endicott Monitoring Program to determine the effects of the Endicott development on nearshore water quality and fish, Snow Goose and caribou populations; various Prudhoe Bay bird studies; Boulder Patch studies; eider use of artificial gravel structures in the nearshore Beaufort Sea; and terrestrial habitat studies. Working as a field researcher and at report preparation for the Arctophila Revegetation Feasibility Study, a program on aquatic plant establishment and distribution in arctic ponds and lakes and the Gravel Vegetation Project, a 10 year study of vegetating gravel pad and roads. Full time with UAF.

7/85-6/87: Environmental Consultant, Anchorage, Alaska. Majority of the work has been with Standard Alaska Production Company as field liaison and consultant for North Slope environmental programs. Observing and participating in field sampling programs on the North Slope, reviewing the reports, providing comments and input on results. Field programs worked on include; the Endicott Environmental Monitoring Program, the Prudhoe Bay Bird Study, and the Boulder Patch Monitoring Program. Have provided review, comment and support for various North Slope environmental studies and attended meetings and conferences with agencies, industry, and contractors on North Slope programs for Standard. Have also assisted in Request for Proposal preparation for several North Slope Oilfield programs and provided review and comment on subsequent contractor selection. Supervisors: Deb Slaybaugh (1985, 1986), Pam Pope (1987) and Chris Herlugson (1988) for Standard.

05/82-7/85. Environmental Scientist/Technical Editor/Engineering Technician for Alaska Environmental Control Services, Inc., 1200 W. 33rd Avenue, Anchorage, Alaska 99503. Responsible for editing engineering reports and editing and production of Environmental Impact Assessments for the firm. Also responsible for researching, writing, and production of Oil Spill Plans, Coast Guard Operations Manuals, and other technical writing commissions. Familiar with oil spill contaminants, regulations, and cleanup problems. Tested water quality of Municipal landfill monitoring wells, assisted in developing the monitoring program to meet state and federal regulations. Sampled ground and surface water for contaminants;

performing field water quality tests as well as securing samples for laboratory analysis. Tested private, commercial, and community wells and septic systems for compliance with municipality, state, and federal regulations. Regularly worked with the public in solving problems relating to the above. No immediate supervisor, owner was Dr. Lee Reid.

4/83-08/83. Environmental Scientist for Sohio Alaska Petroleum Company, Anchorage, Alaska. Environmental consulting for various studies on the North Slope; including the effects of drilling mud effluents and oil spills on aquatic environments. Responsible for reading and commenting on various engineering reports, Environmental Impact Statements, and other documents with an environmental content. Wrote the environmental portion of an exploration plan. Also attended meetings and reported on the development of the Bristol Bay Coastal Management Plan. Gathered, photographed, and identified plants at Prudhoe Bay. Supervisor: Rick Schafer.

06/79-09/81. Researcher at the University of California, Davis, California 99516. Part of a team of engineers and biologists evaluating the feasibility of utilizing marshes and marsh vegetation for wastewater treatment systems. Planted experimental systems and monitored to determine wastewater reduction by different marsh species. Researched, read and annotated some 2000 articles on aquatic vascular plants, then compiled them into an extensive bibliography which was published in 1981. Supervisor: Dr. Marian Stephenson.

12/80-06/81. Consultant to Petaluma Regulatory Wastewater Treatment Facility, Petaluma, California. Assisted in the development of a project utilizing aquatic plants as wind-breaks in oxidation pond settling basins.

09/80-12/80. Consultant to Ecoenergetics, Inc., Vallejo, California. Produced a report on the feasibility of utilizing aquatic vascular plants to de-water sludge of a particular composition.

10/76-06/79. Worked as a full-charge bookkeeper for several small firms, a firm of CPA's, and a temporary bookkeeping service. Searching for a position in my field but unable to locate one in the area, was married at the time.

04/76-09/76. Lab Technician at the University of California, Davis, California 99516. Developed and wrote procedures for monitoring river delta benthic and planktonic community changes in response to water quality parameters and management practices. Supervisor: Dr. Allen Knight, Water Science and Engineering.

07/75-03/76. Worked with freshwater prawns at the Davis Aquaculture Center, Davis, California, 99516. Regulated and tested water quality for prawns from hatching and early growth in salt water to adult growth in freshwater.

01/75-06/75. Lab Technician at the University of California, Davis, California 99516. Responsible for converting and analyzing plant pigment data, producing a report. Supervisor: Dr. Bayer, Botany.

10/73-12/74. Research Assistant at the University of California, Davis, California 99516 while completing a Master's degree. Developed a sampling program to determine the effects of irrigation return water on river periphyton communities. Participated in a two-season sampling program, performed water quality sampling and field and lab tests to determine the effects of agricultural return water on plant growth. Responsible for reports on the project and writing and producing procedures for sampling and analyzing the data. Utilized data from the project to produce a Master's thesis. Supervisor: Dr. Allen Knight, Water Science and Engineering.

PUBLICATIONS AND REPORTS

Alaska Environmental Control Services Inc. 1982. Environmental Assessment for Approval of Industrial Development Lease of Native-Owned Lands. Unalaska Island, Alaska. 92 p.

Stephenson, J.J., G. Turner, P. Pope, J. Colt, A. Knight, and G. Tchobanoglous. 1981. The use and Potential of Aquatic Species for Wastewater Treatment. Appendix A. The Environmental Requirements of Aquatic Plants. Publication No. 65. California State Water Resources Control Board. Sacramento, California. 665 p.

Turner, G. 1984. Spill Prevention Control and Countermeasure Plan for City of Nikolai Fuel Storage Facility, Nikolai, Alaska. Prepared by Alaska Environmental Control Services Inc., Anchorage, Alaska for City of Nikolai, Alaska. 72 p.

Turner, G. 1983. Spill Prevention Control and Countermeasure Plan for Port Lekanoff Facility, Captains Bay, Unalaska Island, Alaska. Prepared by Alaska Environmental Control Services Inc., Anchorage, Alaska. 62 p.

Turner, G. 1983. U. S. Coast Guard Operations Manual for Port Lekanoff Facility, Captains Bay, Unalaska Island, Alaska. Prepared by Alaska Environmental Control Services Inc., Anchorage, Alaska. 72 p.

Turner, G. 1980. Feasibility of Utilizing Aquatic Vascular Plants to Dewater Organic Sludge. Prepared for Ecoenergetics, Inc. Vallejo, California. 22 p.

Turner, G. 1975. The Effect of Agricultural Return Water on Periphyton Communities in the Sacramento River at Knights Landing, California. M.S. Thesis. University of California, Davis, California. 162 p.

NORTH SLOPE PROGRAM EXPERIENCE:

Field

Endicott Environmental Monitoring Program - 1985 through 1990
(Caribou, Snow Geese, Oceanography, Fish, River Discharge, etc.)

Endicott NPDES Monitoring Program - 1987
(Sedimentation, Erosion, benthos)
Endicott Boulder Patch Study - 1986 through 1990
Arctophila Revegetation Feasibility Study - 1987 through 1990
Prudhoe Bay Bird Studies - 1986 through 1990
Gravel Vegetation Study - 1989, 1990
Habitat Studies - 1989, 1990

Report Preparation, Reading, Editing and/or Commenting

For all field programs worked on.
Lisburne Monitoring Studies
ANWR 1002 Report
Industry ANWR Issue Papers
Waterflood draft and final CMR
Niakuk EA and Monitoring Program
Beaufort Sea Fisheries Synthesis
Beaufort Sea Causeway Issues
North Slope Bioaccumulation Study
Habitat Studies

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QUALITY ASSURANCE PROJECT PLAN
for
Feasibility of Restoring the
Bay of Isles and Tonsina Bay in
Prince William Sound and the Gulf of Alaska

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QUALITY ASSURANCE PROJECT PLAN
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4.0 INTRODUCTION

A quality assurance project plan (QAPP) is designed to ensure that all environmentally-related data collected will meet project data quality objectives (DQOs), and be scientifically sound, legally defensible, and of known and documented quality. This plan follows the guidance for preparing QAPPs provided by the Environmental Protection Agency's (EPA's) Quality Assurance Management Staff in the document "Interim Guidelines and Specifications for Preparing Quality Assurance Project Plans (US EPA, 1980). QAPPs are considered to be stand-alone documents that fully explain the methods and activities to be implemented for data collection. Analytical methods and standard operating procedures (SOPs) are included as appendices to the QAPP.

In March 1989 the *Exxon Valdez* ran aground on Bligh Reef in Prince William Sound, spilling approximately 11 million gallons of Prudhoe Bay Crude Oil into the water and impacting over one thousand miles of coastal resources in the Prince William Sound (PWS) and the Gulf of Alaska (GOA). The *Exxon Valdez* oil spill (EVOS) affected the region's environmental habitat, including both floral and faunal populations, as well as recreational, educational, and aesthetic attributes.

According to Gundlach and Hayes (in Ganning et al., 1984), tidal marshes have been classified as the most sensitive shore-type to oil pollution. It has been estimated that 2-20 years are required for tidal marshes to recover naturally (Cairns and Buikema, 1984; RPWG, 1990a; 1990b). Oil is rapidly buried in marshes because they are low energy systems, and degradation is limited under the anaerobic conditions found in these environments (Cairns and Buikema, 1984).

Natural marsh recovery begins when oil toxicity is reduced to a point that can be tolerated by recolonizers (Baker et al., 1990a; 1990b). Full tidal marsh recovery hinges on reduction in oil toxicity; availability of propagules; stability of sediments; and biotic interactions (Getter et al., 1984). Restoration activities in heavily oiled marshes may be expected to require both substantial effort and extended time periods. The presence of oil in high concentrations at a site may complicate restoration efforts, and regrowth in these areas may occur slowly, if at all.

There are two tidal marshes, the Bay of Isles and Tonsina Bay, that are still heavily oiled and lacking in vegetation. A qualitative survey conducted over 4 days in August of 1990 noted (1) heavy effects from residual oil (approximately one acre of mixed Carex and Triglochin) and suspected effects to 1/4 to 1/2 acre of Zostera at the Bay of Isles, and (2) extensive effects to Puccinellia from residual oil at Tonsina Bay; Glaux at higher elevations in this marsh was not affected. Due to these effects on the marshes from the oil, the Bay of Isles and Tonsina Bay require restoration. See Attachment A of the project proposal for maps of Prince William Sound and the Gulf of Alaska, as well as detailed maps of the Bay of Isles and Tonsina Bay showing oiled areas as based on the oil spill maps.

The goal of this feasibility study is to determine whether or not vegetation can be enhanced and/or re-established at the Bay of Isles and Tonsina Bay restoration sites, two tidal marshes known to be heavily impacted by oil.

5.0 PROJECT DESCRIPTION

The project has the following objectives:

- o to quantitatively determine the degree of revegetation success (proportional survival/plot) and relate to crude oil degradation patterns using spatial analysis techniques,
- o at the Bay of Isles test site, increase the rate of ground water discharge to a small section of the wetland in order to determine if sediment characteristics improve more rapidly where the rate of ground water discharge has been increased than in areas where the rate of ground water discharge is normal,
- o to demonstrate whether revegetation success rates improve in areas where the rate of ground water discharge has been increased than in areas where the rate of ground water discharge remains normal.
- o to monitor overall site revegetation success on an annual basis. On plots where there is no revegetation success, replant at similar densities in the succeeding year(s).

Site Restoration

Site restoration will consist of identifying donor sites, conducting restoration trials, and conducting site monitoring. The first annual planting will occur in the spring of 1991. Stands will be established using species native to Prince William Sound and the Gulf of Alaska. At each site, twenty-four 10 m² rectangular plots will be delineated, marked with rebar and revegetated with

nine plantings/m². Broome et al. (1986) discovered that Spartina alterniflora planted 45-60 cm apart were more successful than if spaced farther apart.

Ground Water Flushing

In terms of the hydrological component of the project to be undertaken at the Bay of Isles test site, determine the rate at which water will infiltrate the soil just above the wetland. This can be done by using falling-head or constant-head permeameters (Novitzki 1976). Next, estimate the rate of flow in the nearby stream at the time of the field visit, and compare that to a nearby long-term streamflow record in order to estimate the approximate low flow expected in the stream during the period of the study (Novitzki 1979). Use these data to determine the amount of water available from the stream for creating recharge and the size of impound area necessary to allow the water to infiltrate. Use pipe or flexible hose to divert water from a nearby stream, at an elevation five to ten feet above the mean high water shore line of the wetland, and transport the water to a small, shallow impoundment at the edge of the wetland.

The stream end of the pipe will be anchored at a protected location in a pool, preferably just upstream of a rock riffle. The intake point should be five to ten feet above the elevation of the edge of the wetland to allow water to flow by gravity to the recharge site. The inlet will be protected by a screen or grate to allow a reasonable intake of water for extended periods without maintenance. The pipe or hose will be anchored along its length or buried slightly below grade if possible for protection. No effort will be made to protect the system from freezing because increasing recharge (and consequently discharge) during the warm months will be adequate to demonstrate the effectiveness of this technique.

The recharge area will be created by shoveling loose soil and gravel to make a berm 12 to 18 inches high. The soil will be shoveled from the uphill side so that the soil removal area and berm together form a shallow basin. The basin should be at least 25 but no more than 100 feet long, and from 5 to 10 feet wide. The location of the outlet end of the pipe will be moved up and down hill (at the edge of the recharge area) until the flow rate approximates the desired recharge rate. The flow will be measured volumetrically, using a calibrated container and stop watch.

Project Schedule

Approximately 15 days work will be required at the Bay of

Isles' and Tonsina Bay for initial site delineation, characterization, and planting in the spring of 1991, with 5 days of follow-up monitoring in the early fall of 1991. This estimate is based on the effort of a 5-person field crew (eight to ten hour workdays) for the initial planting and a 2- person crew for the monitoring phases. If all plots exhibit plant survival, future activities will be limited to monitoring restoration success and will require 5 days of field work in the early spring and 5 days in the fall for approximately 4 years following installation. Additional time will be required to collect material and replant any plots on which no plants survived. Care will be taken not to injure sites with equipment or foot traffic. Restoration activities conducted under this project will not interfere with ongoing projects in Prince William Sound and the Gulf of Alaska.

Identification of Donor Site

Field observations in the summer of 1990 have identified several potential donor sites (transplant sources for restoration) for the Bay of Isles and Tonsina Bay:

- 1) Tidal marsh at the head of Outside Bay on Naked Island,
- 2) Tidal marsh on Crafton Island,
- 3) East Bay tidal marsh on Perry Island,
- 4) Culross Passage on Culross Island,
- 5) Tonsina Bay, and
- 6) Fringe tidal marsh around the Bay of Isles and Marsh Bay on Knight Island.

Although these sites may have potential as donor sites, they have not been investigated in detail. Therefore, the following information will be collected and used as criteria to evaluate potential donor sites:

- Species present - The composition of a tidal marsh will factor into its potential to serve as a donor site, based on the species requiring replacement at the Bay of Isles and Tonsina Bay. The site must also have an abundant supply of the appropriate species for revegetation of the Bay of Isles and Tonsina Bay.
- Oil impact - A donor site must be an "unstressed" system (void of unnatural perturbations outside of natural stress), and therefore lacking in any apparent impact from oil.
- Historical treatment record - Again, since a donor site must be "unstressed" relative to the Bay of Isles and Tonsina Bay, a potential donor must not have been

* Dependent on obtaining permission from adjacent land owner.

subjected to any type of treatment or cleanup operations.

- Vigor - To qualify as a donor site, a tidal marsh must exhibit nearly 100% cover of healthy vegetation, again demonstrating the importance of an "unstressed" system.
- Proximity of vegetative donor site - It is cost-effective and ecologically prudent to choose a donor site in close proximity to the Bay of Isles and Tonsina Bay.
- Size of donor plot - The donor site must be large enough that collection of plants for transplantation will not adversely affect the donor. Less than 1 percent cover will be removed from each donor site.

Revegetation

Plants will be installed within 48 hours after being collected. Several alternatives are available for revegetation, including: seeding, bare root, and plugs. Revegetative techniques for Carex involve collecting bare root plants from donor sites, bundling them in groups of 3, and replanting as soon after collection as is feasible. For Puccinellia, it has been shown that plugs survive and grow better than sprigs, so plugs will be used for site restoration (Seneca et al., 1982). When using Puccinellia transplants for restoration, it is important to sufficiently drain the plants.

All transplanted materials will be fertilized at the time of installation. According to Broome (1989) transplants usually benefit from fertilizer the first growing season. Either slow release or conventional water soluble fertilizers can be used. The most widely used fertilization method is approximately 15-30g per plant of slow release Osmocote fertilizer (14-14-14 analysis with a 3 month longevity) in the planting hole (Broome, 1989).

Fertilization was shown to greatly increase growth in Zostera marina in a study by Orth (1977). Fertilizer was massaged by hand into the sediment at the beginning of the experiment (repeated twice) and resulted in a large increase in leaf growth. Studies of tidal marshes affected by the Amoco Cadiz oil spill by Seneca et al. (1982) indicated fertilization (using Mag-Amp and Osmocote) was necessary for significant plant growth because cleanup operations had left large areas void of vegetative cover. Seneca et al. (1982) also observed higher cover for fertilized Puccinellia transplants. Fertilization needs are site-specific, however, and may not be necessary for establishing transplants. Broome et al. (1986) did not use fertilizers and succeeded in establishing a marsh (Spartina alterniflora) for at least 10 years. Fertilizer will be applied once at the outset of the project.

Site Restoration Activities

The following activities will be conducted for site restoration at the Bay of Isles and Tonsina Bay:

- (1) A site description will be provided by completing a Tidal Marsh Restoration Data Form (see Attachment 1), including notations of the amount of oil originally estimated to be present in the area, according to the oil spill maps (i.e., light, moderate, heavy).
- (2) Twenty-four 10 m² rectangular plots, will be placed within each marsh and a reference point will be established. All locations and dimensions will be noted on the data sheet.
- (3) The species to be replaced will be listed, and the total number of required transplants calculated. The amount of fertilizer needed based on the total number of transplants required will be determined.
- (4) A permanent reference point within each treatment plot and will be established and soil samples collected, including replicates (number to be statistically determined), using a random number table to choose the sample points. It will important to note the elevation. A 6.5 cm diameter piston corer will be used to place the sediment into solvent rinsed foil, the sediment will be wrapped, and stored (Burns and Teal, 1979). Labels will be placed on each sample and code with a unique I.D. number assigned. Tape over the label will ensure that the label adheres to the sample and does not smear. Samples will be put into an insulated cooler and transported to the laboratory for analyses. All sampling and analytical utensils contacting the sample will be rinsed with redistilled solvents before use (Burns and Teal, 1979).
- (5) The soil samples will be analyzed for organic content, available nutrients (Ca, Mg, K, P, N as ammonium and nitrate), pH, and salinity at the Soil Science Lab at Oregon State University. Total hydrocarbon and weathered hydrocarbon fractions will be analyzed by SAIC Inc., in San Diego. It will be important to relate revegetation success (survival) to particular oil fractions present.
- (6) Photodocumentation of the site will be made pre- and post-planting and recorded in a log book noting the film frame and roll number.
- (7) Determine an appropriate donor site for both study sites.

- (8) For transplantation of upper tidal marsh vegetation, the methods specified in Appendix 1 will be employed for bare root transplants (based on Spartina alterniflora using Broome, 1989):

Site Monitoring

The Bay of Isles and Tonsina Bay will require monitoring on an annual basis, at the end of the growing season in the fall of 1991, and the spring and fall of subsequent years. Monitoring results should be recorded on the Example Tidal Marsh Site Monitoring Data Form (see Attachment 2).

Ground Water Flushing Activities

Flow from the pipe will be measured several times during the spring field exercise, and once each subsequent visit. The size of the recharge area will be calculated from measurements made after the flow first begins and the pond size stabilizes, near the end of the spring field period, and once each subsequent visit. These two measurements will provide a calculation of the infiltration rate of the soil under prolonged recharge conditions which can be compared to infiltration rates calculated by the falling-head permeameters.

Other measurements will be those sediment characteristics which were planned to be measured at revegetation sites. If sediment characteristics improve most, and revegetation is most successful, near the recharge site, and improvement diminishes proportionally away from that site, there will be a clear demonstration that increased ground water discharge has accelerated sediment cleansing.

6.0 PROJECT QA ORGANIZATION AND RESPONSIBILITIES

A flowchart is particularly useful to show the QA organization of a project and to identify lines of project responsibility for each task or group of measures. Project QA organization is documented in Figure 1. The co-principal investigators (PIs) will be part of the 5-person field crew and will assume responsibility for carrying out the research tasks to ensure quality of the results generated. The Co-PIs will be primarily concerned with the QC aspects of the project. Key QA/QC responsibilities are:

QA Responsibilities

- o participate in the preparation of the QA project plan,
- o ensure that all project participants read and follow the QA project plan,
- o negotiate quality requirements with project officer,

- o train field and analytical staff to perform and evaluate QC measurements,
- o verify that QC activities are performed and data quality is determined as required in the QA project plan, and
- o document QC outputs.

QC Responsibilities

- o follow instrument manufacturer's specifications,
- o perform and document preventive maintenance,
- o maintain up-to-date laboratory notebooks,
- o follow and document deviations from established procedures/methods,
- o make data quality determinations based on QC data collected, and
- o report all problems and corrective actions to the project officer

The project officer is ultimately responsible for the performance and coordination of a specific project. The project officer is management's principal contact regarding the research project. The project officer determines the quality criteria on the basis of intended use of the results to be generated and communicates those criteria to the research staff. Key QA/QC responsibilities are:

QA Responsibilities

- o ensure the development of the QA project plan,
- o ensure that SOPs are developed, review and approve SOPs,
- o negotiate quality requirements with research staff,
- o ensure that required corrective actions are implemented, and
- o review project QC outputs

QC Responsibilities

- o review field logbooks,
- o arrange for performance evaluation or audit samples (when applicable),
- o assist in scheduling audits, and
- o report data quality problems to QA officer

The branch chief is responsible for all projects within a research area and for ensuring that all technical outputs meet the quality requirements of the Laboratory and the Agency. Key QA responsibilities include:

- o review and evaluate work on QA implementation and progress,
- o evaluate QA/QC costs,
- o review and evaluate the quality of outputs generated by each project,
- o review and evaluate audit and performance evaluation reports (when applicable, ensure that corrective actions are implemented), and

- o develop and maintain QA-related communications channels.

Analytical laboratory staff will be required to read this QAPP and agree to comply with the program by completing the Agreement to Comply Form provided in Figure 2.

7.0 OBJECTIVES FOR MEASUREMENT

It is the responsibility of the project officer to define the intended use of the data and to develop, in cooperation with the data users, the DQOs appropriate to the project within the time and resource constraints of the effort. Data quality objectives are described in terms of precision, accuracy, completeness, representativeness, and comparability for all variables to be measured in this project. Development of DQOS must include the following steps:

- o define with specificity the hypothesis, question, or objective to be addressed.
- o establish guidelines for the types and quality of data needed to answer the hypothesis, question, or objective.
- o explain in quantitative terms the possible errors that may arise during the measurement process.

The QA objectives for precision and accuracy for each measure (Table 1) are provided in Table 2. The method of assessing precision and accuracy using different types of quality control (QC) samples is indicated. Completeness is defined as "a measure of the amount of valid data actually obtained from a measurement process required to achieve a particular statistical level of confidence in the data compared to amount expected." The objective for completeness for this pilot project is 85%. The experimental design of this project described in Section 5 is intended to ensure that samples will be collected for oil fraction analysis that are representative of the population to be sampled. The plant transplant aspect of this project does not claim to be representative of all oil contaminated wetland sites because of the pilot-project nature of this study. There is no mandate for demonstrating comparability with other EPA or non-EPA programs for this project. However, it should be a general goal for all projects to collect data that is comparable to other data collected in this scientific field.

8.0 SAMPLING PROCEDURES

The sampling procedures used for all measurements in this project are presented in Table 3 and Appendix 1. The discussion of how sampling locations will be chosen, collection of representative samples, and sample labelling have been provided in Section 5 of this document. Table 3 provides the requirements for sample containers; sample preservation, handling and storage; and recommended holding time limits.

9.0 SAMPLE CUSTODY

Legal sample custody as required by the National Enforcement Investigations Center (NEIC) (US EPA, 1985) is not necessary for this pilot, research activity. Sample transport and handling requirements are provided in Table 3. The laboratory analyzing soil samples is located at Oregon State University, Soil Science Lab in Corvallis, Oregon. Sample collection and labelling will be documented in a field sampling logbook and a daily inventory list of all samples collected will be compiled and checked against the samples at the end of each day. Sample labels will contain site locations, data of collection, name or initials of sample collector and the type of sample (sediment, soil) will be identified. Samples will be shipped to the two analytical laboratories with an inventory list. Verification of sample receipt and evaluation of sample condition upon receipt will be documented by the analytical laboratory. Samples will be stored securely within the analytical laboratory's sample storage area at 40C. Remaining sample will be archived until analyses are completed and results are verified and validated in a secure location, clearly labelled and easily retrievable. The laboratory will track the date of sample analysis and verify that samples were analyzed within recommended holding time limits specified in Table 3.

10. CALIBRATION PROCEDURES AND FREQUENCY

When observational measures are made by more than one person, it is important to address comparability between or among observers. Calibration will include training observers by reviewing the criteria for visual observation and assessment of the condition of transplants after planting to assess viability and in evaluating viability over time. Remeasurement by all observers of 10% of the total quadrants or plots measured will be used to calibrate visual observations and provide a numerical index of variability among observers.

For analytical variables (elemental analyses) the number of standards used, their composition, and concentration will be documented by the analytical laboratory. The sample pattern will be documented to ensure that all QC samples are analyzed as required. Either high and low concentration QC check samples or certified reference standards will be used to ensure calibration

accuracy during batch sample analysis. Low concentration check samples or certified reference materials will be used to verify batch-to-batch detection limits and as an indirect method to monitor daily detection limits. The results from the analysis of at least 7 low concentration check samples are used to calculate a standard deviation. The method detection limit is the Student's T value for a one-tailed test at the 99% confidence level with n-1 degrees of freedom. It will be necessary to identify quality control check samples (QCCSs) that were used to indicate the need for recalibration as a required corrective action.

Balances used in this project will be calibrated annually under a service contract with a competent firm specializing in balance calibration and maintenance. Annual calibration will be verified by a sticker attached directly to the balance.

Project pH meters will be calibrated before use using two calibration standards bracketing the normal operating range. The calibration will be verified using a quality control check sample. Meter calibration should also be verified at the end of the analysis period.

11. ANALYTICAL PROCEDURES

Table 1 lists the methods to be used in this study. Standard and published methods are provided whenever possible. Methods for the determination of hydrocarbon fractions and weathering to be performed by SAIC are provided in Appendix 2. Methods for analyses to be performed by Oregon State University's Soil Testing Laboratory are provided in Appendix 3.

12. DATA REDUCTION, VALIDATION, AND REPORTING

Sample collection from the field can be traced by entries in the field sampling logbook, the inventory list, and the sample receipt log. Results from the analytical laboratory will be documented in both hard copy and database format on floppy disk. Raw data sheets (specifying reporting units) will be retained by the analytical laboratory. The data format required for computer file entry should be provided to the analytical laboratory. The analytical laboratory is expected to verify data entry accuracy (by visual or electronic checking procedures) of 100% of the entries. Summary statistics such as range and reasonableness checks will be used to identify outlier and error values. Data files will be backed-up regularly. Statistical tests used in final data reports will be clearly identified.

13. INTERNAL QUALITY CONTROL CHECKS

Internal QC activities ensure the quality of the data collected by verifying the precision and accuracy of analytical results in comparison to the data quality objectives specified in

Table 2. Internal QC checks also ensure that instruments are operating properly and the calibration curves are valid as sample analysis proceeds. The frequency of performing QC activities was not decided for all analytical project cooperators at the time this plan was prepared. The required frequency of the QC activities specified in Table 2 and defining the appropriate warning and control limits, and the associated corrective actions required when control limits are exceeded will be part of the contractual agreements with the participating analytical laboratories.

14. PERFORMANCE AND SYSTEM AUDITS

The QA staff of the Environmental Research Laboratory-Corvallis perform a technical systems audit (TSA) or data quality audit (DQA) of all projects. TSAs are conducted prior to or concurrent with initial data collection activities to:

- o familiarize project staff with EPA QA requirements and procedures,
- o evaluate the implementation of the QA activities specified in the QA project plan (QAPP), and
- o provide assistance in attaining the objective to collect data of known and documented quality.

Long-term projects are audited every two years or at the request of the project officer. A data quality audit (DQA) is an evaluation of the documentation associated with data quality indicators of measurement data to verify that the data are of known quality. The primary purpose of this type of audit is to verify the availability of quantitative and qualitative indicators of data quality. Availability of data quality indicators depends upon the proper collection, interpretation, and reporting of information required to characterize the quality of the data.

This project is considered a pilot study. During the first year of the project no TSAs will be conducted by ERL-C QA staff. Instead a DQA will be conducted at the conclusion of data collection activities to ensure that data meet project DQOs. However, Region 10 QA staff may elect to review either field or analytical laboratory activities.

The analytical laboratory at OSU in the Department of Soil Science will determine the organic content, nutrients, pH and salinity in soil/sediment samples. The laboratory will be required to follow this QA plan and provide the QC data specified in Table 2. This laboratory has a QA program in place which is described in Appendix 3B.

The routine QA activities practiced by SAIC are included in appendix 2.

A performance audit (PA) is a quantitative evaluation of a measurement system involving a challenge to the system by the use

of reference samples of known composition and concentration. PAs are used to determine whether a measurement system is operating within established control limits at the time of the audit. This provides an objective assessment, in terms of precision and accuracy, of the data being generated by the system. These data may be compared to control limits established for the system (or DQOs) to identify out-of-control conditions. The results of the audit also are used to verify the accuracy and precision of data being generated in routine QC analyses conducted for the measurement system. Availability of appropriate standard or certified reference materials for the analysis of oil fractions will be investigated. This material will be analyzed in replicate in every sample batch to evaluate precision and accuracy both within and between batches.

15. PREVENTIVE MAINTENANCE

Routine preventive maintenance of all field equipment analytical instruments listed in Table 1 will be performed when QC checks indicate the need for maintenance or when dictated by routine maintenance schedules.

16. SPECIFIC ROUTINE PROCEDURES USED TO ASSESS DATA PRECISION, ACCURACY, AND COMPLETENESS

Precision and accuracy are evaluated using the approaches specified in Table 2. Precision is defined as a measure of scatter among independent repeated observations or measures of the same property under prescribed conditions. Precision is usually expressed in terms of the standard deviation as:

$$s = \left(\sum_{i=1}^n (X_i - \bar{X})^2 / (n-1) \right)^{1/2}$$

where \bar{X} is the mean of n measurements and X_i is the value of the i th measurement. Accuracy is defined as the degree to which a measured value agrees with a "true" or accepted value (or a calculated mean or median). Measures of precision and accuracy are to be completed when QC samples are analyzed and will be summarized and submitted with final data reports. Control charts will be encouraged to be used to routinely monitor precision and accuracy in the participating analytical laboratories.

17. CORRECTIVE ACTIONS

Corrective actions are performed when QC check samples indicate analytical problems, audits identify concerns, or when routine preventive maintenance indicates a problem. Table 6 provides an example of corrective actions required for an atomic absorption spectrophotometer when precision and accuracy goals are

not attained. All required corrective actions will be implemented as soon as a problem is identified. These actions will be documented and provided to the EPA project officer and Co-PIs. If required corrective actions affect data quality, the specific affected samples, observations, or other data should be explicitly identified. Caveats limiting the use of these data may be necessary when reporting final project results. All participating analytical laboratories will develop corrective action logbooks. Corrective actions required in the field can be documented in the project field notebook or on data sheets.

18. QUALITY ASSURANCE REPORTS (TO MANAGEMENT)

As discussed in Section 16, measures of precision and accuracy are to be completed when QC samples are analyzed and will be summarized and submitted with final data reports. Control charts will be encouraged to be used to routinely monitor precision and accuracy in the participating analytical laboratories. When corrective actions are required, the action taken and the results of the action can be discussed and documented in the final data report along with any problems that may affect the quality of the data or limit the use of the data.

Project deliverables and a schedule for their completion should be agreed upon between the principal investigators, the EPA project officer and all project participants. Turn-around times for analysis of soil samples and receipt of laboratory results should be clearly stated in any formal or informal agreements.

19. REFERENCES

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Table 1. Project Activities and Approach

Activity	Equipment/ Approach	Method/ Reference
Donor Site Activities	plastic bags shovel metal coat hangers shovels twist-tie fasteners construction paper containers	Upper Tidal Marsh (Broome, 1989) Subtidal Marsh (Fonseca et al., 1982) and (Fonseca, 1989)
Revegetation	coring device dibble dive knife fertilizer metal coat hangers shovels twist-tie fasteners	
Site Restoration		
site description biomass percent cover vigor	Tidal Marsh Restoration Data Form (Attachment 1)	(Simensted et al. 1989)
estimate of oil spill damage	Oil Spill Maps	
plot establishment	measuring tape (m),	
soil/sediment sample	piston corer, compass	
salinity	conductivity meter	
site photodocumentation pre-planting post-planting	camera, film, logbook	
Annual monitoring visual assessment of survival and vigor	Tidal Marsh Site Monitoring Data Form (Attachment 2)	
For <i>Puccinellia</i> (eelgrass): density, number of shoots/ unit area	Quadrat method	(Simensted et al. 1989)

Table 1. Project Activities and Approach
(continued)

Activity	Equipment/ Approach	Method/ Reference
<u>SOIL/SEDIMENT ANALYSIS</u>		
<u>SAIC, San Diego, CA</u> hydrocarbon fraction	Methylene chloride extraction, Fluorsil column clean-up, hexane partitioning, evaporate to dryness, gravimetric analysis	Appendix 2
Hydrocarbon fraction weathering	capillary guard column	
<u>Oregon State University</u> nutrients	1 N ammonium acetate extraction, atomic absorption spectrophotometer (Perkin- Elmer 372)	Appendix 3
Ca, Mg, K		
phosphorus	Bray's solution extraction (if pH <7.0), molybdate blue method w/ in-line dialyzer, continuous flow analyzer, (Alpkem) (if pH > 7.0 extract with sodium bicarbonate)	
ammonium-N	KCl extraction, indophenol blue method, continuous flow analyzer, (Alpkem)	
nitrate-N	KCl extraction, Cd reduction, continuous flow analyzer, (Alpkem)	
organic content	ground to pass 0.5 mm sieve, Walkley Black titration	
soil pH	2:1 (Water:Soil) Electrode/meter	

Table 2. Objectives for Data Quality

Activity	Data Quality Parameter Evaluated
For analysis of soil/sediment samples for nutrients:	
analysis of "control" samples from site where plants were collected	Provides a blank value, verifies site was un- contaminated
analysis of low concentration QC check sample analysis of high concentration QC check sample	calibration verification, detection limit verification (from replicate results)
analysis of replicate samples	Evaluates sample precision (within 10%)
analysis of 3 replicates of certified reference standard/ batch	Evaluates method accuracy (within 10%) Evaluates method precision (within 10%)
analysis of low concentration spike sample analysis of high concentration spike sample	detection limit verification, estimation of method % recovery
For pH:	
analysis of replicate samples	Evaluates sample precision (within 5%)
analysis of 3 replicates of certified reference standard/ batch	Evaluates method accuracy (within 5%) Evaluates method precision (within 5%)
For hydrocarbon fraction, and organic content:	
analysis of replicate samples	Evaluates sample precision (within 10-15%)

Table 3. Sample Collection, Handling, and Preservation

Sample Type	Sample Container	Preservation Method/ Storage	Minimum Sample Size	Maximum Holding Time
TRANSPLANTS				
Upper Tidal Marsh Vegetation				
Carex plants	3/plastic bag	keep cool, in shade, moist	---	overnight
Puccinellia plants	plugs	keep cool, in shade, moist	---	overnight
Subtidal Marsh Vegetation				
Zostera marina	15 shoots/ clump	anchor w/ coathanger, construction paper, twist- tie in water- filled container	---	overnight
soil/ sediment	glass or plastic, cap tightly	store and transport on ice or cool	2 l.<2mm. fraction	as soon as possible after sample reaches room temperature

ATTACHMENT 1
Tidal Marsh Restoration Data Form

EXAMPLE TIDAL MARSH RESTORATION DATA FORM

Investigators Name _____
 Date _____
 Time _____
 Location _____
 Segment Number/ID _____

Restoration Assessment

<p>1 Site Description: _____</p>	<p>2 Extent of original oiling (Based on oil map) _____</p> <p>_____ moderate Areal percent _____</p> <p>_____ heavy</p>																												
<p>3 Extent of living/dead vegetation:</p> <p>i) Apparent Cover: _____%</p> <p>ii) _____% aboveground biomass</p> <p>iii) Belowground biomass present beyond limit of aboveground biomass? _____ Y _____ N</p> <p>if yes, extent of total marsh is _____%; location _____</p>	<p>4 i) Location/tidal zone of each treatment area</p> <p>Permanent reference point location (landmark) _____</p> <table style="width:100%; border-collapse: collapse;"> <thead> <tr> <th style="text-align: left;">Distance from reference point (yards)</th> <th style="text-align: left;">Elevation</th> </tr> </thead> <tbody> <tr><td>#1T _____</td><td>#1T _____</td></tr> <tr><td>#2T _____</td><td>#2T _____</td></tr> <tr><td>#3T _____</td><td>#3T _____</td></tr> <tr><td>#4T _____</td><td>#4T _____</td></tr> <tr><td>#5T _____</td><td>#5T _____</td></tr> <tr><td>#6T _____</td><td>#6T _____</td></tr> </tbody> </table> <p>ii) substrate type (S=Sand, SH=Shale, R=Rock/Cobble, M=Mud)</p> <table style="width:100%; border-collapse: collapse;"> <tbody> <tr><td>#1T _____</td><td>#4T _____</td></tr> <tr><td>#2T _____</td><td>#5T _____</td></tr> <tr><td>#3T _____</td><td>#6T _____</td></tr> </tbody> </table> <p>iii) Location/tidal zone of each control area</p> <table style="width:100%; border-collapse: collapse;"> <thead> <tr> <th style="text-align: left;">Distance from reference point (yards)</th> <th style="text-align: left;">Elevation</th> </tr> </thead> <tbody> <tr><td>#1C _____</td><td>#1C _____</td></tr> <tr><td>#2C _____</td><td>#2C _____</td></tr> <tr><td>#3C _____</td><td>#3C _____</td></tr> </tbody> </table> <p>iv) Salinity _____‰</p>	Distance from reference point (yards)	Elevation	#1T _____	#1T _____	#2T _____	#2T _____	#3T _____	#3T _____	#4T _____	#4T _____	#5T _____	#5T _____	#6T _____	#6T _____	#1T _____	#4T _____	#2T _____	#5T _____	#3T _____	#6T _____	Distance from reference point (yards)	Elevation	#1C _____	#1C _____	#2C _____	#2C _____	#3C _____	#3C _____
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<p>5 Approximate area (m²) to be restored per species:</p> <table style="width:100%; border-collapse: collapse;"> <thead> <tr> <th style="text-align: left;">List: species</th> <th style="text-align: left;">area</th> </tr> </thead> <tbody> <tr><td>_____</td><td>_____</td></tr> <tr><td>_____</td><td>_____</td></tr> <tr><td>_____</td><td>_____</td></tr> </tbody> </table>	List: species	area	_____	_____	_____	_____	_____	_____	<p>6 Number of transplants needed: 9 holes/m² @ 3 plants/hole</p> <table style="width:100%; border-collapse: collapse;"> <thead> <tr> <th style="text-align: left;">species</th> <th style="text-align: left;">number</th> </tr> </thead> <tbody> <tr><td>i) _____</td><td>_____</td></tr> <tr><td>ii) _____</td><td>_____</td></tr> <tr><td>iii) _____</td><td>_____</td></tr> <tr><td>iv) TOTAL _____</td><td>_____</td></tr> </tbody> </table>	species	number	i) _____	_____	ii) _____	_____	iii) _____	_____	iv) TOTAL _____	_____										
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<p>7 Amount of fertilizer needed: _____ lbs.</p> <p># of plants (6 iv) x .066 lbs. = lbs. of fertilizer needed</p>	<p>8 Vegetative Donor Site:</p> <p>i) Proximity to restoration site (approximate miles) _____</p> <p>ii) Size (m²) _____</p> <p>iii) Donor site identification number _____</p>																												
<p>10 Soil Analyses (record shipping information on reverse side): Soil sample taken? _____ Y _____ N</p> <p>i) If yes, number of samples (including duplicates) _____</p> <p>ii) I.D. numbers _____</p> <p>iii) Method of storage _____</p>	<p>9 Comments: _____</p> <p>11 Oil characteristics at the site:</p> <p>i) surface _____</p> <p>ii) subsurface _____</p> <p>iii) asphalt _____</p> <p>iv) sheen _____</p>																												

ATTACHMENT 2
Tidal Marsh Site Monitoring Data Form

EXAMPLE TIDAL MARSH SITE MONITORING DATA FORM

Investigators Name _____
 Date _____
 Time _____
 Location _____
 Segment Number/ID _____

<p>1 Restoration method used:</p> <p>i) _____ fertilization</p> <p>ii) _____ transplant/fertilize</p> <p>iii) Date treated _____</p>	<p>2 Species used for each treatment plot:</p> <p>List:</p> <p>#1T _____ #4T _____ #2T _____ #5T _____ #3T _____ #6T _____</p>																																																												
<p>3 Living/dead vegetation cover per treated and control areas:</p> <p>#1T _____ % #1C _____ % #2T _____ % #2C _____ % #3T _____ % #3C _____ % #4T _____ % #5T _____ % #6T _____ %</p>	<p>4 Substrate samples collected for oil/nutrient analysis (Y or N)</p> <table style="width: 100%; border-collapse: collapse;"> <thead> <tr> <th style="width: 50%;"></th> <th style="width: 25%; text-align: center;">Oil</th> <th style="width: 25%; text-align: center;">Nutrient</th> </tr> </thead> <tbody> <tr><td>#1T</td><td>_____</td><td>_____</td></tr> <tr><td>#2T</td><td>_____</td><td>_____</td></tr> <tr><td>#3T</td><td>_____</td><td>_____</td></tr> <tr><td>#4T</td><td>_____</td><td>_____</td></tr> <tr><td>#5T</td><td>_____</td><td>_____</td></tr> <tr><td>#6T</td><td>_____</td><td>_____</td></tr> </tbody> </table>		Oil	Nutrient	#1T	_____	_____	#2T	_____	_____	#3T	_____	_____	#4T	_____	_____	#5T	_____	_____	#6T	_____	_____																																							
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<p>5 Apparent vigor</p> <table style="width: 100%; border-collapse: collapse;"> <thead> <tr> <th style="width: 30%;"></th> <th style="width: 5%;">1T</th> <th style="width: 5%;">2T</th> <th style="width: 5%;">3T</th> <th style="width: 5%;">4T</th> <th style="width: 5%;">5T</th> <th style="width: 5%;">6T</th> <th style="width: 5%;">1C</th> <th style="width: 5%;">2C</th> <th style="width: 5%;">3C</th> </tr> </thead> <tbody> <tr> <td>i) Vigorous¹ (%)</td> <td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td> </tr> <tr> <td>ii) Healthy² (%)</td> <td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td> </tr> <tr> <td>iii) Low³ (%)</td> <td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td> </tr> <tr> <td>iv) Poor⁴ (%)</td> <td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td> </tr> <tr> <td>v) Dying⁵ (%)</td> <td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td><td><input type="text"/></td> </tr> </tbody> </table>			1T	2T	3T	4T	5T	6T	1C	2C	3C	i) Vigorous ¹ (%)	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	ii) Healthy ² (%)	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	iii) Low ³ (%)	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	iv) Poor ⁴ (%)	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	v) Dying ⁵ (%)	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>
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iii) Low ³ (%)	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>																																																				
iv) Poor ⁴ (%)	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>																																																				
v) Dying ⁵ (%)	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>																																																				
<p>6 Comments:</p>																																																													

- Key:**
- 1 (healthy color, >80% cover)
 - 2 (healthy color, 20-80% cover)
 - 3 (healthy color, <20% cover)
 - 4 (unhealthy color)
 - 5 (unhealthy color - brown stems; sparse cover)

Appendix 1
Vegetation Transplant

Appendix 1

Vegetation Transplant

For transplantation of upper tidal marsh vegetation, the methods specified below will be employed for bare root transplants (based on Spartina alterniflora using Broome, 1989):

1. Obtain bare root transplants (Carex) from the edges of the selected donor tidal marshes. Loosen the plants with a shovel and remove from the marsh. Carefully remove sediment from the roots and bundle in groups of three. Using a plastic bag, place transplants in the bag so the roots are covered, and keep the roots moist.
2. To hand plant, work in pairs. The first worker creates a hole with a dibble approximately 15 cm deep, and adds 0.21 lb of fertilizer for one bundle per hole. A second worker inserts plants and firms the soil around the plants. For this project, whether there is a need to plant in pairs or individually should be determined in the field.
3. For transplantation of upper tidal marsh vegetation the following methods will be employed for plug transplants:
 - 3a. Obtain plug transplants (Puccinellia) from a donor site by inserting a coring device approximately 20 cm into the substrate, and removing the intact plug from the ground.
 - 3b. Remove plug from the coring device and place in plastic bags to keep the plug moist during transport.
 - 3c. To hand plant, create a hole with a dibble or coring device large enough to hold the plug, insert 0.21 lb of fertilizer into the hole, and insert the plug. Firm the soil around the plug to anchor it.
4. Take a second picture of the site once the transplants have been planted, and mark in a log book the film frame and roll number.
5. Observations involving biomass, percent cover, and vigor will rely on the experience and professional judgement of the investigator.

Appendix 2

Methods from SAIC for
Hydrocarbon Fraction and Weathering Analysis

I. SUMMARY OF METHOD OF SATURATED HYDROCARBON (HC) AND POLYNUCLEAR AROMATIC (PAH) COMPOUNDS ANALYSIS FOR SEAWATER SAMPLES

A. Sample Preservation

Methylene chloride is added to the sample in a volume ratio of 1:10 methylene chloride:seawater.

B. Sample Extraction

The extraction procedure for seawater samples is the application of EPA SW 846 Method 3510 (separatory funnel method).

B.1 Transfer the preserved known volume of seawater sample (10 ml for a shaker experiment and 100 ml for respirometric experiment) into a 250 ml separatory funnel.

B.2 Add 1 ml of 50 ppm HC surrogate standards (o-Terphenyl or n-Decylcyclohexane) and 1 ml of 1 ppm PAH surrogate standards (Naphthalene-c8, Acenaphthene-d10, and Chrysene-d12).

B.3 Rinse the sample bottle with 20 ml methylene chloride and add the extract to the separatory funnel.

B.4 Seal and shake the separatory funnel for 1-2 minutes, with periodic venting to release excess pressure.

B.5 Allow the organic layer to separate from the water phase and collect the methylene chloride extract.

B.6 Repeat the extraction two more times using fresh portions of methylene chloride (30 ml each). The three extracts are passed through an anhydrous sodium sulfate column, and combined in a Kuderna-Denish evaporation concentrator.

B.7 The extract is concentrated to a final volume of 1 ml on the K-D apparatus.

B.8 The extract is now ready for fractionation.

C. Sample Fractionation

C.1 Activate 60/200 -mesh silica gel at 210 C for 24 hours. Prepare a slurry of 8-10 gm of activated silica gel in hexane.

C.2 Place the silica gel slurry into a 10 mm ID x 25 cm long column.

C.3 Tap the column to settle the silica gel and elute the hexane. Add 1 to 2 cm of anhydrous sodium sulfate to the top of the silica gel.

C.4 Drain the column until the solvent is just above the sodium sulfate layer.

C.5 Transfer 1 ml of the sample extract from B.8 onto the column. Just prior to exposure of sodium sulfate to the air, elute the fraction according to the following order

of solvents.

<u>Fraction</u>	<u>Elution amount (ml)</u>	<u>Compound Class</u>
Hexane	15 - 30	Aliphatic hydrocarbon
Hexane:Benzen	45	Aromatic hydrocarbon (1:1)

C.5 Concentrate each fraction into 1 ml using a K-D evaporative concentrator.

D. Aliphatic Hydrocarbon Analysis by GC/FID

D.1 Instrument: Hewlett-Packard 5880A Gas Chromatograph with Flame Ionization detector.

Column: 0.75 mm ID x 30 m long DB-5 with direct injection in the splitless mode.

Operating Parameter:

- Injector port temperature	250° C
- Detector temperature	350° C
- Temperature program	
initial temperature	50° C
hold time	5 minute
program rate	7° C/min
final temperature	300° C
final hold time	35 minute
final run time	75 minute or less
- Injection volume	2 uL
- Carrier gas	He 5 ml/min
- Make up gas	He 20 ml/min
- Detector	Air 240 ml/min

D.2 Calibration Curve

Prepare a five point calibration curve (Table 1) of concentration 10, 50, 100, 200, and 250 ppm in methylene chloride. Each of standard is made up of pristane, phytane, C7 through C25 and C26, C28, C30, C32, C34, C36 and C38. Add to each standard 50 ug of o-terpheyyl surrogate or n-decylcyclohexane surrogate standard and 50 ug x-androstane internal standard per 1 ml of the calibration standard.

D.3 Sample Analysis

Spike 50 ug of x-androstane internal standard into each 1 ml of sample prior to analysis.

II. SUMMARY OF QA/QC PLAN

A. Aliphatic Hydrocarbon QA/QC Protocol

A.1 Relative standard deviation of response factors of five point calibration must be within $\pm 25\%$.

A.2 Daily check standard of 100 ppm is determined each day or for every 10 samples analyzed. The % difference of the response factors must be within $\pm 25\%$.

A.3 Percent recovery for n-decylcyclohexane surrogate should be within 60 - 140%.

A.4 Matrix spike and matrix spike duplicate are analyzed for every 20 samples or with every sample sent - whichever is more frequent. The spike criteria are as follows:

A.4.1 Matrix spike percent recovery of C15, C20, and C28 must be within 60 - 140%.

A.4.2 Only one compound can be below its required minimum percent recovery.

B. Aromatic Hydrocarbon QA/QC Protocol

B.1 GC/MS is tuned to meet PFTBA tuning criteria as shown below for every 12 hours of analysis.

PFTBA tuning criteria:

Mass ion	% Acceptance relative to base peak m/z 69
51	1 - 6
69 (base peak) 100	
131	30 - 50
219	30 - 60
414	1 - 2

B.2 Relative standard deviation of response factors of five point calibration standard must be within 40%.

B.3 Daily check standard is determined every 12 hours of analysis. The % difference of the response factors must be within 35%.

1. MEASUREMENTS FOR OIL CHEMISTRY IN SEDIMENT AND WATER SAMPLES

Routine measurements in sediment samples are made for three variables related to oil chemistry: (1) total residue weight extracted by methylene chloride, (2) total hexane-extractable and non-hexane-extractable residue weights, and (3) hydrocarbon composition and content determined by flame ionization detector-gas chromatography (FID-GC). A flow-diagram of the protocol for analyses of these variables is shown in Figure 1. An example of the information data sheet used by laboratory personnel to record sample extraction information for sediment samples is shown in Figure 2.

Measurements in water samples are made for two variables related to oil chemistry: (1) total residue weight extracted by hexane and (2) hydrocarbon composition and content determined by flame ionization detector-gas chromatography (FID-GC). A flow-diagram of the protocol for analyses of these variables in water samples is shown in Figure 3. An example of the information data sheet used by laboratory personnel to record sample extraction information for water samples is shown in Figure 4.

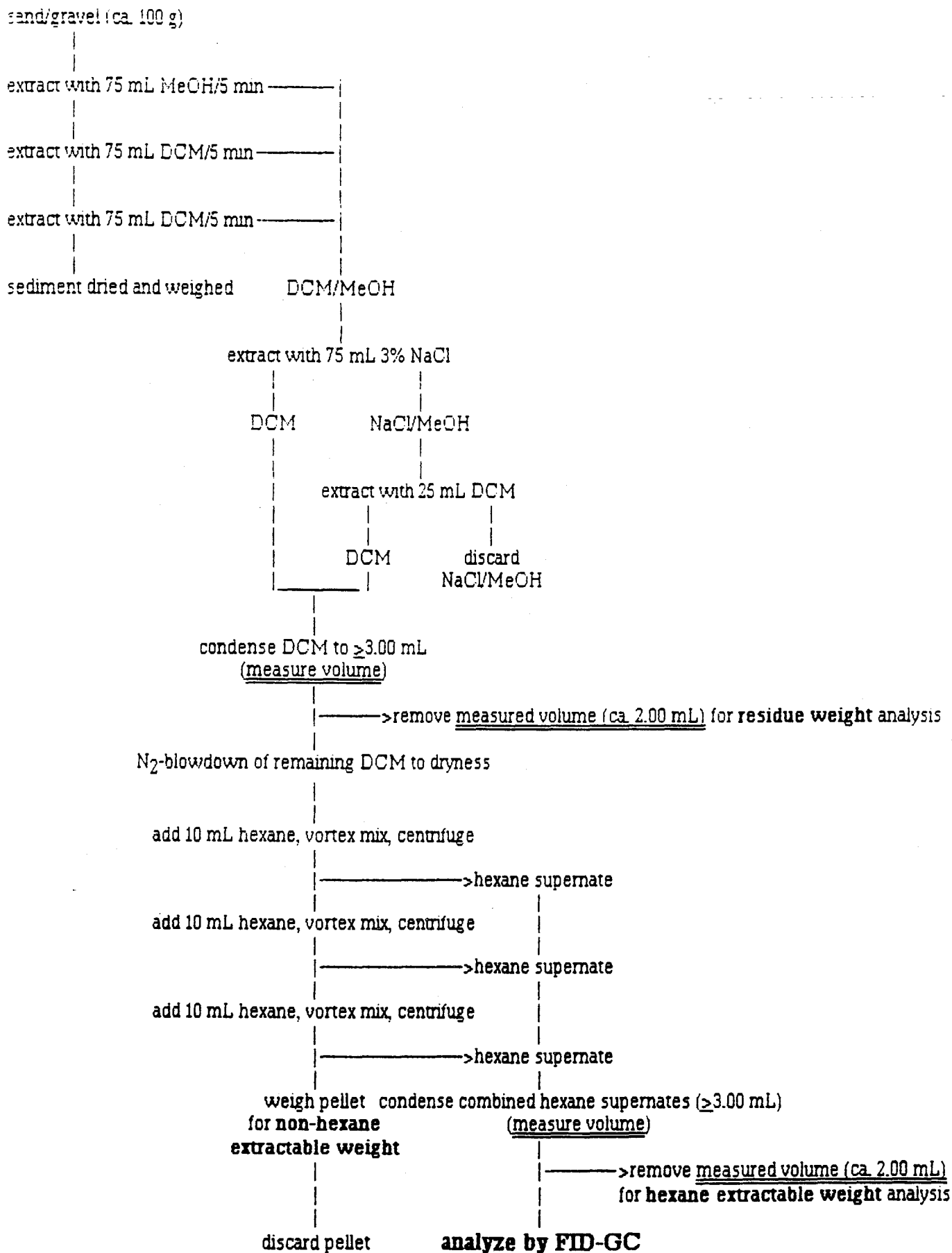
1.1. Total Residue Weight

1.1.1 Sediment samples: methylene chloride extraction/residue weight measurement

Approximately 100 g of a sediment sample in a sand/gravel size range (ca. 4-13 mm diameter) are placed in a clean 250 mL glass Erlenmeyer flask. A volume of 75 mL of pesticide-quality methanol (MeOH) is added and the flask is shaken on a mechanical shaker table for 5 min. The MeOH is then decanted through a paper filter (VWR brand, Grade No. 613) into a 500 mL separatory funnel. The sediment sample is extracted (i.e., shaken for 5 min) with two additional 75 mL volumes of pesticide-quality methylene chloride (DCM). The DCM supernates are also passed through the filter paper into the separatory funnel containing the initial MeOH extract. The weight of the sediment extracted is determined by drying the solvent-extracted sediment in an oven at 45°C, transferring the sediment to a tared weighing pan, and determining the sample weight with a Mettler Model PE160 balance capable of reading to 0.001 g.

A 75 mL volume of a 3% NaCl:freshwater solution (w:w; pre-extracted with DCM) is added to the separatory funnel containing the combined DCM-MeOH extract from the sediment sample. The funnel is then shaken for approximately 1 min, the solvent phases allowed to separate, and the DCM layer transferred to a 1000 mL glass round-bottom flask. The residual water/NaCl/MeOH solution in the separatory funnel is back-extracted (i.e., shaken for ca. 1 min) with 25 mL of DCM. Following solvent phase separation, the latter DCM layer is also transferred to the round-bottom flask. Several Teflon boiling chips are added and a three-ball Snyder column attached to the round-bottom flask. The DCM in the round-bottom is reduced in volume to approximately 5 mL over a 45°C water bath. The condensed DCM is transferred quantitatively (i.e., with DCM rinses) to a graduated cylinder. The volume in the cylinder is adjusted to a measured volume ≥ 3.00 mL under a stream of high purity nitrogen (N₂) gas. A measured aliquot

SAND/GRAVEL SAMPLE EXTRACTION (ca. 100 g extracted)



**EPA/SAIC 1990 Bioremediation Program
Extraction Sheet for Oil Analysis—Sediment Sample
DCM/Hexane-Extractable Procedure**

I. Sample Identification:

SAIC Analysis Identification Number: _____
 Batch Identification Number: _____
 Sample Identification Number: _____
 Miscellaneous Sample Information: _____

II. Sample Extraction/Residue Weight Determination:

a) Extraction Analyst: _____ Date of Extraction: _____

b) Extraction Information:

i) Dry Weight of Sediment Extracted: _____ g (1)

ii) Solvent Extractions:

initial MeOH/DCM extractions:

volume MeOH/agitation time: _____

volume DCM/agitation time: _____

number of DCM extractions: _____

back-extraction:

volume of 3% NaCl/agitation time: _____

volume DCM/agitation time: _____

number of DCM extractions: _____

iii) Final DCM Extract

Final extract volume [A]: _____ mL (2)

Residue weight determination for final extract

volume for residue weight measurement [B]: _____ mL (3)

residue weight measurements:

tare: _____ g

residue + tare measurement #1: _____ g

residue + tare measurement #2: _____ g

measured DCM residue weight [C]: _____ g (4)

total residue weight in final sample extract [C x (A/B)]: _____ g

Residue weight/dry weight of sediment _____ g/g

III. Hexane-Extractable/Non-Hexane-Extractable Weight Determinations:

a) Extraction Analyst: _____ Date of Extraction: _____

b) volume of final DCM extract used for hexane-extraction step [D]: _____ mL (5)

c) hexane extractions:

i) volume hexane/agitation time: _____

number of hexane extractions: _____

ii) final hexane extract

final volume [E]: _____ mL (6)

hexane-extractable and non-hexane-extractable weight determinations:

vol. of fin. extr. taken for hexane-extractable weight measurement [F]: _____ mL (7)

weight measurements:

hex-extractable

non-hex-extractable

tare: _____ g

residue + tare measurement #1: _____ g

residue + tare measurement #2: _____ g

measured weights [G]: _____ g (8)

total weights in sample: {Gx(E/F)x(A/D)} = _____ g {Gx(A/D)} = _____ g

Hexane-extractable weight/total residue weight of sample: _____ %

Non-hexane-extractable weight/total residue weight of sample: _____ %

Hexane-extractable/Non-hexane-extractable ratio: _____

IV. GC Sample Run Information for Hexane-Extractable Fraction:

a) Date GC vial crimped: _____ GC operator: _____

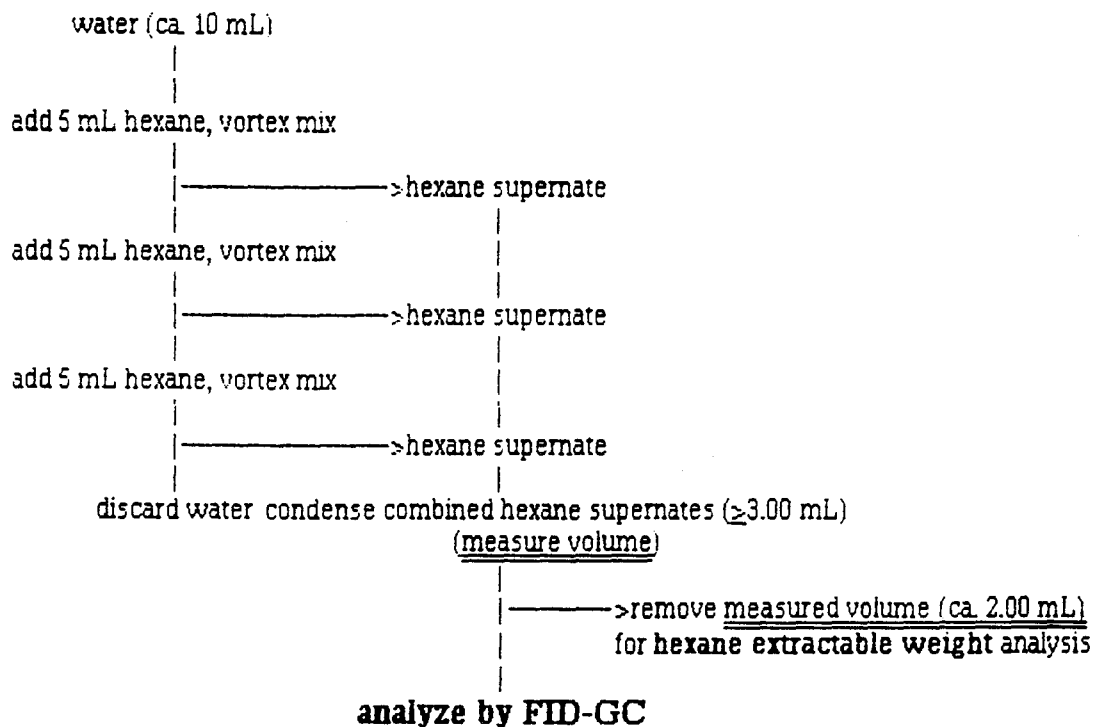
c) HP instrument ID: _____ PIV (uL): _____

d) HP run date: _____ HP run number: _____

g) GC file ID for sample: _____ Dilution factor: _____

V. General Comments:

WATER SAMPLE EXTRACTION (ca. 10 mL extracted)



EPA/SAIC 1990 Bioremediation Program
Extraction Sheet for Oil Analysis—Water Sample
(hexane extraction method)

I. Sample Identification:

SAIC Sample #: _____
Batch ID: _____
Sample Number: _____
Collection Site: _____
Collection Date: _____
Miscellaneous Sample Information: _____

II. Sample Extraction:

a) Extraction Analyst: _____ Date of Extraction: _____

b) Extraction Information:

i) Volume of Water Extracted: _____ mL

ii) Solvent Extractions:

extraction:

volume hexane/agitation time: _____

number of hexane extractions: _____

iii) Final Sample Extract:

Final extract volume [A]: _____ mL

Residue weight determination for final extract:

vol. of fin. extr. taken for res. wt. measurement [B]: _____ mL

residue weight measurements:

tare: _____ g

residue + tare measurement #1: _____ g

residue + tare measurement #2: _____ g

residue + tare measurement #3: _____ g

measured residue weight [C]: _____ g

total residue weight in final sample extract [C*(A/B)]: _____ g

Residue weight/volume of water: _____ mg/L

III. GC Sample Preparation/Run Information:

a) Date vial crimped: _____ GC operator: _____

c) HP instrument ID: _____ IV/PTV (uL/uL): _____

d) HP run date: _____ HP run number: _____

g) GC file ID for sample: _____

h) Comments: _____

(ca. 2.00 mL) is transferred to a tared aluminum weighing pan and allowed to dry at room temperature to a constant weight. Total residue weight for the sample is determined as the difference between the weight measurements for the weighing pan+sample and the pan tare, with appropriate correction being made for the volume fraction of the initial DCM extract used for the weight measurement. Weight measurements for the pan tare and sample+tare are made with a Mettler Model PE100 balance capable of reading to 0.0001 g. The residual DCM fraction of the sample not used for the total residue weight measurement is used for the following hexane-extraction procedure (Section 1.2).

1.1.1 Water samples: hexane extraction/residue weight measurement

A water sample having a 10.0 mL volume is contained in a 20-mL VOA vial equipped with a Teflon-lined screw cap. A 5 mL volume of pesticide-quality hexane is added and the vial is mixed on a Vortex Mixer for ca. 1 min. Following phase separation, the hexane is transferred to a graduated cylinder with either a gas-tight syringe or a disposable Pasteur pipet. This extraction procedure is repeated with two additional 5-mL volumes of hexane, which are also transferred to the graduated cylinder. The hexane volume in the cylinder is adjusted to a measured volume ≥ 3.00 mL under a stream of high purity nitrogen (N_2) gas. A measured aliquot (ca. 2.00 mL) is transferred to a tared aluminum weighing pan and allowed to dry at room temperature to a constant weight. Total residue weight for the sample is determined as the difference between the weight measurements for the weighing pan+sample and the pan tare. Weight measurements for the pan tare and sample+tare are made with a Mettler Model PE100 balance capable of reading to 0.0001 g.

1.2. Hexane-Extractable and Non-Hexane-Extractable Weight: hexane extraction of initial DCM residue fraction—Sediment Samples Only

A measured volume of the condensed DCM extract from Section 1.1.1 is reduced to dryness in a glass tube under a stream of high purity nitrogen (N_2) gas. A 10 mL volume of pesticide-quality hexane is added to the tube and mixed with a Vortex Mixer for ca. 1 min. The tube is then centrifuged to precipitate the "non-hexane-extractable" fraction, and the "hexane-extractable" supernate is transferred to a clean glass tube. The hexane (10 mL) vortexing/centrifugation step is repeated two additional times. All hexane supernates (i.e., the "hexane-extractable" fraction) are combined in the glass tube and reduced in volume to approximately 2-4 mL under a stream of N_2 gas. The extract is then transferred quantitatively (i.e., with hexane rinses) to a 10 mL graduated cylinder and the exact volume recorded. A measured aliquot (ca. 2.00 mL) of the extract is then transferred to a tared aluminum weighing pan and allowed to dry at room temperature. Total hexane-extractable weight for the sample is determined from the extract weight in the pan, with appropriate corrections being made for the sample volume fractions used for weight measurements in this section and Section 1.1.1. Total non-hexane-extractable weight for the sample is determined by allowing the hexane-extraction tube to dry (i.e., after removal of the final hexane supernate), determining its weight, removing all of the non-hexane-extractable pellet with DCM rinses, drying the tube again, and measuring its final weight. The non-hexane-extractable weight for the sample is determined from the difference

in the tube weights with and without the non-hexane-extractable pellet, with appropriate corrections being made for the volume of the initial sample extract removed for the total residue weight measurement (Section 1.1.1). Weight measurements for the hexane-extractable and non-hexane-extractable weight determinations are made with a Mettler Model PE100 balance capable of reading to 0.0001 g.

1.3. Measurement of hydrocarbon composition and content by FID-GC analysis

Analysis of sample extracts for hydrocarbon composition and content are performed on a Hewlett-Packard (HP) 5890 gas chromatograph (GC) equipped with a flame ionization detector (FID), a HP 7673A autosampler and controller, an 82169A HP-IL/HP-IB Interface, a HP 3396A integrator, and a HP 9133H disk drive. A fused silica capillary chromatography column in the GC is used for compound separations. GC operating and temperature programming parameters used for analyses are:

- GC column: DB-5 liquid phase, 30 m length X 0.32 mm ID, 0.25 μ m film thickness (J&W Scientific, 91 Blue Ravine Road, Folsom, CA 95630)
- GC injection mode: splitless, 1 min valve closure
- GC injection temperature: 285°C
- GC detector temperature: 350°C
- GC oven programming rate:
 - initial temperature: 45°C, 5 min hold
 - temperature ramp: 3.5°C/min
 - final temperature: 280°C, 20 min hold
- sample injection volume: 1.0 μ L

Quantitation for hydrocarbons is accomplished with an external standard method. The standard solutions consist of aliphatic hydrocarbons containing a sequential mixture of n-alkanes with even and odd numbers of carbon atoms (n-C8 through n-C30 plus n-C32) and the isoprenoid compounds pristane and phytane. Integration of peaks in all chromatograms (i.e., standards and samples) is accomplished with valley-to-valley baseline placement.

A mixture of polynuclear aromatic hydrocarbons (PAHs) consisting of compounds with two to six ring structures was also used in an attempt to identify PAH compounds in sample extracts. PAH compounds could not be reliably identified in the FID-GC chromatograms of sample extracts in the absence of a physical separation of aliphatic and aromatic fractions for sample extracts. Hence, identification and quantitation of PAH compounds will require additional treatment of the sample extracts (e.g., physical separation of aliphatic and aromatic fractions and/or gas chromatography-mass spectrometry analysis).

1.3.1. Standard solutions for FID-GC analyses

Table 1

n-ALKANE STANDARD SOLUTIONS FOR FID-GC

Compound	conc in primary standard (ng/uL)	concentrations of aliphatics in working standard solutions (ng/uL)		
		[125]	[25]	[5]
nC-8	244	122	24.4	4.88
nC-9	260	130	26.0	5.20
nC-10	248	124	24.8	4.96
nC-11	248	124	24.8	4.96
nC-12	260	130	26.0	5.20
nC-13	272	136	27.2	5.44
nC-14	260	130	26.0	5.20
nC-15	272	136	27.2	5.44
nC-16	268	134	26.8	5.36
nC-17	320	160	32.0	6.40
pristane	268	134	26.8	5.36
nC-18	248	124	24.8	4.96
phytane	256	128	25.6	5.12
nC-19	252	126	25.2	5.04
nC-20	432	216	43.2	8.64
nC-21	248	124	24.8	4.96
nC-22	276	138	27.6	5.52
nC-23	320	160	32.0	6.40
nC-24	244	122	24.4	4.88
nC-25	260	130	26.0	5.20
nC-26	252	126	25.2	5.04
nC-27	244	122	24.4	4.88
nC-28	244	122	24.4	4.88
nC-29	256	128	25.6	5.12
nC-30	240	120	24.0	4.80
nC-32	240	120	24.0	4.80

NOTE: all standards in 100% hexane

Table 2

PAH STANDARD SOLUTION FOR FID-GC

Compound	concentrations of PAHs (ng/uL)
naphthalene	66.2
2-methylnaphthalene	65.0
1-methylnaphthalene	67.5
biphenyl	66.2
2,6-dimethylnaphthalene	66.2
acenaphthylene	63.6
acenaphthene	68.6
2,3,5-trimethylnaphthalene	59.1
fluorene	65.5
phenanthrene	66.1
anthracene	50.1
1-methylphenanthrene	65.5
fluoranthene	65.9
pyrene	66.1
benz[a]anthracene	56.8
chrysene	66.1
benzo[b]fluoranthene	65.9
benzo[k]fluoranthene	65.9
benzo[e]pyrene	66.2
benzo[a]pyrene	59.6
perylene	49.8
indeno[1,2,3-c,d]pyrene	58.6
dibenz[a,h]anthracene	49.9
benzo[ghi]perylene	58.6

NOTE: standard in 100% toluene; obtained from NIST

The primary standard for aliphatic hydrocarbons is prepared by combining known quantities of neat n-alkane (n-C8 through n-C30 plus n-C32) and isoprenoid (pristane and phytane) compounds in a volumetric flask and bringing to volume with hexane. Nominal concentrations for compounds are 250 ng/uL, with specific concentrations being shown in Table 1. Working solutions of the standards are prepared at three concentration levels for FID-GC analyses by appropriate dilutions of the primary standard with hexane. Nominal concentration levels of individual compounds in the working standards are 125 ng/uL, 25 ng/uL, and 5.0 ng/uL, with specific concentrations being shown in Table 1.

The standard used in attempts to identify PAH compounds in sample extracts is a certified standard solution prepared by NIST (i.e., the National Institute of Standards and Technology, Gaithersburg, MD). Compound concentrations in this standard are approximately 60 ng/uL, with specific concentrations being shown in Table 2.

1.3.2. Initial stability calibration of the FID-GC

Before sample extracts can be analyzed for their hydrocarbon content and composition, the FID-GC must meet specified initial instrument stability calibration criteria. These calibration criteria are performed with injections of the three working aliphatic standard solutions (i.e., nominal concentrations of 5, 25, and 125 ng/uL for compounds). Peaks for compounds in chromatograms of the standard solutions must be 90% resolved. Peak resolution (PR) is calculated with the following formula:

$$PR = [1 - (\text{height of valley between 2 peaks} / \text{height of smaller of 2 peaks})] \times 100.$$

Retention times (RTs) for all identified compounds in the standard solutions must also vary by no more than $\pm 1.0\%$ from the mean RT for the three injections. Finally, response factors (RFs) for aliphatic compounds in the working standards must meet certain reproducibility criteria related to instrument response. RFs are calculated as:

$$RF_x = \text{ng of compound x on column} / \text{GC area counts for compound x}.$$

In the three working solutions for the aliphatic standards, the RFs for nC-17, pristane, nC-18, and phytane must not vary by more than $\pm 25\%$ from the mean value for the three solutions. No more than three of the RFs for all remaining n-alkanes can vary by $> \pm 40\%$ from their respective means for the three standard solutions.

If an injection for one of the three standard solutions for the aliphatic standards is responsible for failure to meet the preceding criteria, the "offending" standard solution can be injected one additional time. If the result of the reanalysis meets the stability criteria, injection of sample extracts can begin. If results of the reanalysis do not meet the stability criteria, sample extracts cannot be run and routine maintenance must be performed on the instrument to correct the problem.

1.3.3. Ongoing calibration of the FID-GC

Following initial instrument calibration, analysis of sample extracts occurs. However, the FID-GC must be recalibrated (i.e., checked for ongoing stability of the instrument response) with an aliphatic standard solution at least once every 24 hours. The standard solution for this ongoing stability calibration is the 25 ng/uL working standard for the aliphatics. RFs for nC-17, pristane, nC-18, and phytane in the ongoing standard calibration check cannot vary by >30% from the means obtained in the initial 3-point calibration (Section 1.3.2.). No more than three of the RFs for all remaining n-alkanes can vary by >40% from their means in the initial 3-point calibration.

If the ongoing calibration check for the aliphatic standard does not meet the preceding instrument stability criteria, the standard solution can be injected one additional time. If the result of the reanalysis meets the stability criteria, injection of sample extracts can continue. If results of the reanalysis do not meet the criteria, a new 3-point initial stabilization procedure must be initiated (Section 1.3.2) and/or routine instrument maintenance on the GC must be performed. All sample extracts injected after the last acceptable calibration check must also be reanalyzed on the FID-GC.

1.3.4. Quantitation for concentrations of identified n-alkane and isoprenoid compounds

N-alkane and isoprenoid (i.e., pristane and phytane) compounds are quantified in FID-GC chromatograms of sample extracts by an external standard method that uses the aliphatic standard solutions used to calibrate the GC. Initial and ongoing instrument stability criteria for the FID-GC must be acceptable (i.e., Sections 1.3.2 and 1.3.3) for quantitation to proceed for sample extracts. For samples, chromatographic peaks are identified as aliphatic compounds by comparison with retention times for specific compounds in the closest preceding injection of the aliphatic standard solution. The retention time (RT) for a peak in a sample chromatogram must be within $\pm 1.0\%$ of the absolute RT for the compound in the standard for assignment of compound identity.

Final concentrations for hydrocarbons identified by FID-GC in sample extracts will be reported in units of mass of a hydrocarbon compound per unit mass of total residue weight (i.e., DCM-extractable residue for sediments, as determined in Section 1.1). Calculation of these hydrocarbon concentrations is done with the formula:

$$C_X = (A_X \times RF_X) \times (\text{vol}_{\text{fin. hex.}} / \text{vol}_{\text{GC inj}}) \times (\text{vol}_{\text{fin. DCM}} / \text{vol}_{\text{for hex ext}}) \times (1 / \text{tot. res. wt.})$$

where

C_X = concentration of analyte x per unit of total residue weight (in g/g),

A_X = FID-GC area counts for analyte x,

RF_X = response factor for analyte x (see Section 1.3.2),

$\text{vol}_{\text{fin. hex.}}$ = total volume of the final hexane-extractable fraction analyzed by FID-GC,

$\text{vol}_{\text{GC inj}}$ = volume of the hexane-extractable fraction injected into the FID-GC,

$vol_{fin.DCM}$ = final volume of the initial DCM extract,

$vol_{for\ hex\ ext}$ = volume of initial DCM extract processed through the hexane-extractable procedure, and

$tot.res.wt.$ = total DCM-extractable residue weight for the sample (in g, see Section 1.1).

Values for RF_x in this calculation are obtained from the closest preceding injection of the mid-level aliphatic (i.e., 25 ng/uL) standard injected into the GC.

1.3.5. Quantitation for concentrations of total resolved peaks and the unresolved complex mixture (UCM) in FID-GC chromatograms

FID-GC chromatograms of sample extracts will normally contain (1) a variety of resolved hydrocarbon peaks including identified n-alkanes, pristane, and phytane as well as other unidentified peaks and (2) an unresolved complex mixture (UCM) that appears as a "hump" above the background chromatogram baseline and beneath the resolved peaks. Aliphatic compounds identified in the resolved peak fraction are quantified as described in Section 1.3.4.

Concentrations for unidentified resolved peaks in a sample extract between two contiguous n-alkanes (exclusive of pristane and phytane) are estimated by summing areas for all resolved peaks between the two n-alkanes and using the following formula:

$$C_y = (A_y \times RF_y) \times (vol_{fin.hex.}/vol_{GC\ inj}) \times (vol_{fin.DCM}/vol_{for\ hex\ ext}) \times (1/tot.res.wt.)$$

where

C_y = estimated concentration for all resolved peaks between two adjacent n-alkanes per unit mass of total residue weight,

A_y = FID-GC area counts for the sum of the resolved peaks between the two adjacent n-alkanes

RF_y = mean of the response factors for the two adjacent n-alkanes (see Section 1.3.2),

$vol_{fin.hex.}$ = total volume of hexane-extractable fraction that was analyzed by FID-GC,

$vol_{GC\ inj}$ = volume of hexane-extractable fraction injected into the FID-GC,

$vol_{fin.DCM}$ = final volume of initial DCM extract,

$vol_{for\ hex\ ext}$ = volume of initial DCM extract used for hexane-extractable measurement,

$tot.res.wt.$ = total DCM-extractable residue weight for sample.

The concentration for the total resolved peaks is calculated as the sum of all identified aliphatics (i.e., n-alkanes plus pristane and phytane; Section 1.3.4) plus the sum of the concentrations for the unidentified resolved peaks between all adjacent n-alkanes as computed with the preceding equation.

Hydrocarbon concentrations for the UCM in a sample extract are estimated by determining the area of the UCM in a FID-GC chromatogram with an electronic digitizing tablet (Kurta Corporation). The estimated concentration for the UCM is calculated as:

$$C_Z = (A_Z \times P_Z \times RF_Z) \times (\text{vol}_{\text{fin.hex.}} / \text{vol}_{\text{GC inj}}) \times (\text{vol}_{\text{fin.DCM}} / \text{vol}_{\text{for hex ext}}) \times (1 / \text{tot.res.wt.})$$

where

C_Z = estimated concentration for the UCM per unit mass of total residue weight,

A_Z = area of the UCM "hump" in the chromatogram in digitizing tablet units (at a specified attenuation)

P_Z = factor for converting digitizing tablet area units to equivalent GC area units (determined for the attenuation specified for A_Z)

RF_Z = mean of the response factors for all n-alkanes between the nC-12 and nC-32 (see Section 1.3.2 for RF determination),

$\text{vol}_{\text{fin.hex.}}$ = total volume of hexane-extractable fraction that was analyzed by FID-GC,

$\text{vol}_{\text{GC inj}}$ = volume of hexane-extractable fraction injected into the FID-GC,

$\text{vol}_{\text{fin.DCM}}$ = final volume of initial DCM extract,

$\text{vol}_{\text{for hex ext}}$ = volume of initial DCM extract used for hexane-extractable measurement,

tot.res.wt. = total DCM-extractable residue weight for sample.

The P_Z factor for the digitizing tablet is determined by comparing areas for two n-alkane peaks (usually nC-15, nC-23, pristane, and/or phytane) in chromatograms for all of the sample extracts obtained by both the digitizing tablet and the GC integrator (the latter being the method used for all resolved peaks in chromatograms). The overall mean of these values from all of the sample chromatograms was used as the P_Z value. Digitizing tablet area units are dependent on the attenuation at which a chromatogram is run, whereas area counts determined by the electronic integrator attached to the GC are not. Hence, the P_Z factor for converting digitizing tablet area units to equivalent GC area units for UCM determinations is dependent on the attenuation used for a particular chromatogram.

1.4. QC (Quality Control) procedures for measurements related to oil chemistry

1.4.1. Method blanks

Analytical method blanks involve analysis of solvent blanks through the analytical procedures illustrated in Figures 1 and 3. Method blanks are analyzed with a frequency of at least 1 for every 12 field samples.

1.4.2. QCCS (Quality Control Check Samples)

A QCCS involves analysis of a known weight of unweathered Prudhoe Bay crude oil through the analytical procedures illustrated in Figures 1 and 3. QCCS samples consist of a weighed amount of unweathered Prudhoe Bay crude oil (ca. 100 mg). Data for repeated analyses of QCCS samples should remain relatively constant, and the data are used to control and assess precision and accuracy. Variables monitored in QCCS samples include (1) total residue weight (as a percentage of the initial crude oil weight used for the QCCS sample), (2) total hexane-extractable and non-hexane-extractable weights (as percentages of the initial crude oil weight), (3) concentrations of individual n-alkanes, pristane, phytane, total GC-resolved peaks, and the UCM per unit mass of the initial oil, and (4) concentration ratios for nC-17/pristane, nC-18/phytane, and total GC-resolved peaks/UCM. Control charts will be developed and monitored for these variables in QCCS samples during the course of analyses. QCCS samples will be analyzed with a frequency of at least 1 for every 12 field samples during routine oil analyses.

The ideal material for the QCCS analyses would be a naturally weathered North Slope crude oil. However, sufficient quantities of such a material are not readily available. Therefore, unweathered Prudhoe Bay crude is used for the QCCS. When using unweathered crude, it must be recognized that volatile components (e.g., lower molecular weight compounds) will be lost during certain sample treatment steps (e.g., the drying step to determine total residue weight). However, these lower molecular weight components will already be absent from oil obtained from field samples due to evaporation and dissolution processes that have previously affected the oil. Effects of evaporation losses of more volatile components on overall weights for unweathered Prudhoe Bay crude oil have been investigated, with results being illustrated in Figure 5. Data for the figure were generated by putting 280-300 mg of unweathered Prudhoe Bay crude oil in weighing pans, maintaining the pans open to the atmosphere at room temperature (70-80°C), and taking pan weights over time. Results in the figure indicate that total residue weight recoveries for QCCS samples of unweathered Prudhoe Bay crude oil should be on the order of only 75% following drying of the residue extract in a drying pan.

1.4.3. Analytical triplicates/duplicates

Triplicate or duplicate subsamples of selected field samples will be analyzed to evaluate "within batch" variability during laboratory analyses. Precision for these analyses are calculated as relative standard deviations (RSD) for triplicates or relative percent differences (RPDs) for duplicate analyses of samples. RSD as a percent is calculated as:

$$\text{RSD} = (\text{standard deviation}/\text{mean}) \times 100.$$

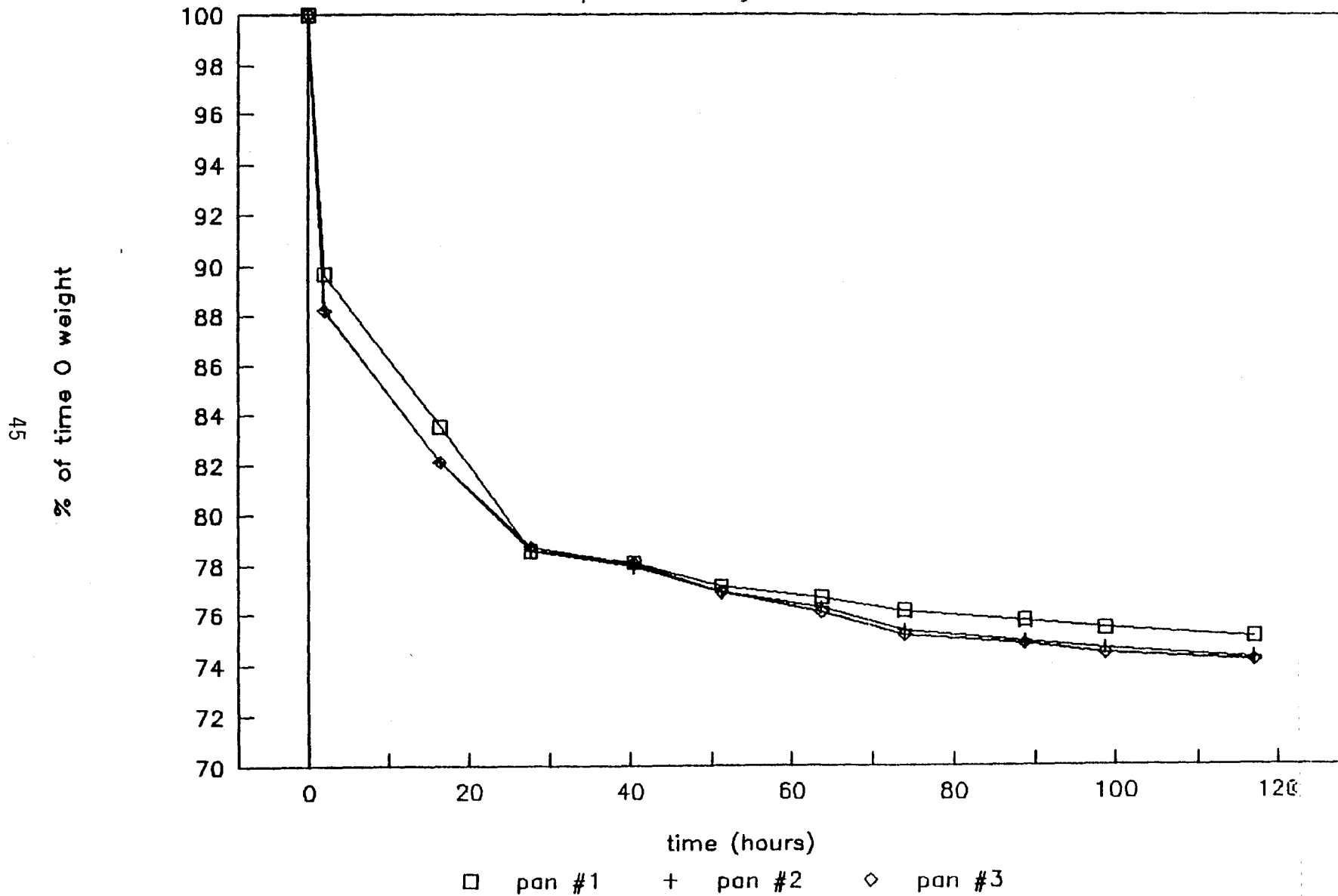
RPD as a percent is calculated as:

$$\text{RPD} = [(x_1 - x_2)/x_{\text{ave}}] \times 100$$

where x_1 and x_2 are measured values for two analyses and x_{ave} is the mean of the two analyses. The frequency of the triplicate/duplicate analyses is according to decisions made by the EPA

Unweathered Prudhoe Bay Crude Weights

evaporation study---June 1990



Principal Scientist (H. Pritchard) and/or the project data management team (e.g., D. Heggem, A. Neale, D. Chaloud).

1.4.4. QAX (Quality Control Check for Extraction Efficiency)

To estimate extraction efficiency for oil from sediment samples, a second extraction (i.e., a QAX) for selected samples will be performed using the protocols described in Sections 1.1, 1.2, and 1.3. The QAX extract will be processed in an identical manner to the initial sample extract. Results of the QAX analysis will be taken to represent complete extraction of oil from samples if values for measured variables (e.g., total residue weight, hexane-extractable and non-hexane-extractable weights, and/or FID-GC measured concentrations for hydrocarbons) in the QAX are <15% of values measured in the initial extraction of the sample.

1.5. Detection limits

1.5.1. Sediment dry weight

Dry weights for sediments extracted for oil chemistry are measured with a Mettler Model PE160 balance capable of reading to 0.001 g. Sediment samples extracted for this program will be in the range of 100 g dry weight.

1.5.2. Total (DCM-extractable) residue weights and hexane-extractable and non-hexane-extractable weights

Measurements for both the total (i.e., DCM-extractable) residue weight (Section 1.1.1) and hexane-extractable and non-hexane-extractable weights (Section 1.2) for sediment samples are measured with a Mettler Model AE100 balance capable of reading to 0.0001 g.

1.5.3. Hydrocarbon compound concentrations by FID-GC

The HP5890 FID-GC (see Section 1.3) is capable of detecting approximately 0.0001 ug (i.e., 1×10^{-10} g) of an individual n-alkane compound injected into the GC. Using the formula in Section 1.3.4 for sediment samples, this yields a detection limit of approximately 1 ug/g of total residue weight for an individual n-alkane compound in a sediment sample with the following assumptions:

- (1) a final sample hexane volume of 4.00 mL,
- (2) an FID-GC sample injection volume of 1.00 uL,
- (3) a final DCM extraction volume of 4.00 mL,
- (4) 2.00 mL of the DCM volume used for the hexane extraction procedure, and
- (5) a total DCM-extractable residue weight for a sample of 0.8 g (i.e., a residue amount frequently measured in extractions of 100 g of sediment from field samples for the 1990 Bioremediation Program).

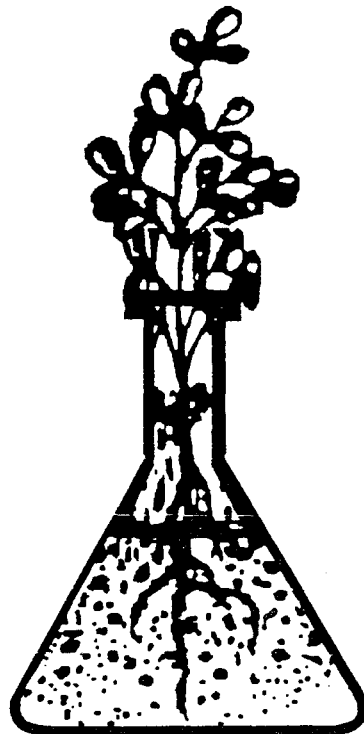
Specific detection limits for n-alkane compounds will vary between samples because items 1, 3, 4, and 5 in the preceding assumptions will vary between samples.

Appendix 3A

Oregon State University, Soil Testing Laboratory Methods

**Methods of Soil Analysis Used in the
Soil Testing Laboratory
at Oregon State University**

D. A. Horneck, J. M. Hart, K. Topper, B. Koepsell



Methods of Soil Analysis Used in the Soil Testing Laboratory at Oregon State University

D. A. Horneck, J. M. Hart, K. Topper, B. Koepsell*

INTRODUCTION

General

This manual describes and documents procedures used in the Oregon State University Soil Testing Laboratory (OSUSTL), and to supply information on the appropriate documentation of these methods. Of the numerous methods for soil analysis, research at Oregon State University indicates that the procedures outlined in this publication are suitable for Oregon conditions.

The Cascade Mountain Range is a natural boundary that separates Oregon into eastern and western sectors. Western Oregon soils tend to be acidic, while the soils in eastern Oregon tend to be slightly acidic or alkaline. In view of these differences, some testing procedures differ for eastern and western Oregon. For example, the phosphorus test for western Oregon requires a dilute acid-fluoride (Bray P1) extraction solution, while sodium bicarbonate is used for samples from eastern Oregon.

Although reference is made to specific scientific supplies and instruments used in the OSUSTL, similar equipment from other manufacturers can be substituted. Mention of a model or brand name is neither an endorsement nor a promotion for the product.

The appendix contains a combination of alternate procedures, seldom used procedures and instructions for standardization of an acid.

Future Considerations

Improving analytical procedures for fertilizer recommendation is an on-going project at the College of Agricultural Sciences, Ag. Experiment Station, Extension Service, Department of Soil Science and the OSUSTL. Consequently, after thorough research, soil testing procedures and methods of reporting are periodically updated. Comments from the farming and university communities, along with suggestions from the fertilizer industry, commercial laboratories, and agriculture consultants are considered. Future topics for research include:

1. Using a volume scoop for routine analyses versus weighing samples.
2. Evaluating a universal extractant, such as Melich III for analyses performed on an ICP.
3. Computerizing of data acquisition from laboratory equipment.

*Donald A. Horneck, research assistant, and John Hart, Extension soil scientist, Oregon State University. Karl Topper, research assistant, Utah State University; formerly research assistant, Oregon State University. Barbara Koepsell, lab technician, Oregon State University.

4. Rewriting the computer program which prints and writes fertilizer recommendations.
5. Compile an annual report that includes data from other soil testing laboratories.

When major analytical changes are accepted, an updated edition of this publication would be made available.

Collection and Preparation of Soil Samples

Collecting soil samples from the field is an integral part of soil testing. Samples must represent the soil in the field from which it is taken. This involves obtaining 20-40 subsamples per sample submitted for analysis. Information on soil sampling is provided in Oregon State University Extension Circular 628, "How to Take a Soil Sample and Why." Sampling instructions are also available at county Extension offices or from OSUSTL.

Samples should be submitted in a standard soil sample bag or in a plastic container. Plastic containers are preferable to metal containers for collecting and mixing soil samples. Contamination may be a problem for boron (B) and zinc (Zn) when samples are collected and stored in certain kinds of paper bags. In the field, extreme care is necessary to avoid contaminating the soil sample with fertilizer or with extraneous materials from the sampling tools.

When the soil samples arrive at the OSUSTL, they are placed on trays and dried in a forced-air drying cabinet at 35 C or lower. Drying at higher temperatures may affect analytical results. Soil samples normally dry in 24 to 48 hours and are then pulverized and sieved with a Custom Laboratory Equipment Co. Dynacrush soil crusher.¹ Soil passing through the 14-mesh (2 mm) stainless steel sieve is returned to the original sample bag and stored for analysis. OSUSTL releases soil test results and fertilizer recommendations immediately after sample analysis has been completed. Soil samples are stored for future reference for 4 to 6 months, then discarded.

Accuracy and Precision

Laboratory instruments are calibrated using standard solutions that are either purchased commercially or mixed by the OSUSTL. Standard soil samples are also maintained as reference samples for evaluating

Documentation of Methods

The analytical methods used in the OSUSTL, including appropriate literature citations, are outlined in the following sections. Modifications of the published methods with respect to changes in reagents or in procedural detail is described under "Comments." Some procedures have been modified to facilitate the use of a continuous-flow analyzer. Since this equipment is not available in all laboratories, alternative procedures are also reported. A general reference for procedures used in analyzing soil is *Methods of Soil Analysis* published by the American Society of Agronomy in Madison, Wisconsin (1982).

Use of ppm

In this manual the use of parts per million (ppm) is meant to be equivalent to milligrams per liter (mg/L) or milligrams per kilogram (mg/kg), with the weight of 1 liter of water equal to 1000 g or 1 kg. General use of ppm follows:

$\text{ppm} = \text{mg/L} = \text{mg L}^{-1}$
for solids weighed in water

$\text{ppm} = \text{mg/kg} = \text{mg kg}^{-1}$
for results on a dry weight basis

ACKNOWLEDGEMENTS

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ANALYTICAL METHODS

pH 1:2 soil to water ratio

A. Reagents

Buffer solutions for calibration of pH meter.

Note: The buffer solutions can be purchased if desired.

1. pH 4.005 - 0.05 M potassium biphthalate ($\text{KHC}_8\text{H}_4\text{O}_4$). Dry $\text{KHC}_8\text{H}_4\text{O}_4$ for two hours at 110 C. Dissolve 10.21 g $\text{KHC}_8\text{H}_4\text{O}_4$ in distilled water and dilute the solution to a volume of 1 L with distilled water. As a preservative, add 1.0 mL chloroform or a crystal (about 10 mm in diameter) of thymol per liter of the buffer solution.
2. pH 6.860 - 0.025 M KH_2PO_4 and 0.025 M Na_2HPO_4 . Dry the two phosphate salts for two hours at 110 C. Dissolve 3.40 g of KH_2PO_4 and 3.55 g of Na_2HPO_4 in distilled water and dilute the solution to a volume of 1 L with distilled water. As a preservative, add 1.0 mL of chloroform or a crystal (about 10 mm in diameter) thymol per liter of the buffer solution.
3. pH 9.177 - 0.01 M $\text{Na}_2\text{B}_4\text{O}_7 \cdot 10\text{H}_2\text{O}$. Dry the $\text{Na}_2\text{B}_4\text{O}_7 \cdot 10\text{H}_2\text{O}$ for two hours at 110C. Dissolve 3.81 g in distilled water and dilute the solution to 1 L.
4. Hydrochloric acid, 0.1 N HCl - Dilute 8.3 mL of concentrated HCl to 1 L volume with distilled water.

B. Procedure

1. Scoop 20 cc (g) of dry soil into a 3-oz paper cup or 100 mL beaker.
2. Add 40 mL of distilled water and stir thoroughly.
3. Let stand about 15 min, stir a second time, and allow suspended soil to settle for at least 15 min before reading pH.
4. Calibrate the pH meter according to instrument instructions using two of the prepared buffer solutions. After instrument calibration, rinse the electrodes with 0.1 N HCl and then distilled water to remove any trace of the buffer solutions.
5. Read the pH by placing the electrodes in the supernatant liquid and swirling gently. Record the pH to the nearest 0.1 unit.
6. Rinse the electrodes with distilled water and pat dry between pH determinations.
7. When the meter is not in use, immerse the electrodes in pH 6.860 buffer.
8. pH readings should be made routinely on known standard soil samples, every 15 samples in the OSUSTL.

C. Comments

This method is described by McLean (1982). The one used has a 1:2 soil-water ratio where the pH is measured in the supernatant instead of in the soil suspension, for convenience and to minimize the errors introduced by liquid junction potential.

Buffer solutions should be prepared fresh at least once a month. If solutions are purchased, expiration dates need to

be noted. The pH meter needs to be calibrated periodically when making a series of determinations to check for drift. Check samples should also be incorporated into a series of analyses to ensure accurate readings. For pH measurements in soil a combination (single) or a dual electrode can be used. The OSUSTL uses a dual electrode.

Greweling and Peech (1968) indicate that pH may shift slightly with a change in the soil-to-water ratio used in sample preparation. Seasonal fluctuations in pH can also be expected. Soil pH will tend to be lower for samples collected after heavy fertilization. Conversely, pH may increase as the concentration of fertilizer salts decreases. Salt accumulation in soil tends to lower pH, and salt removal by leaching will have the opposite effect of raising pH. Fluctuations in pH due to seasonal or analytical effects may vary from 0.1 to 1.0 pH units.

Soil pH can also be determined using prepared salt solutions; this indicates the effect of salts in the sample. For example, the pH value obtained using 1 N KCl will normally be 1 to 1.5 units lower than the distilled water value. The soil pH measured in 0.01 M CaCl_2 will be about 0.4 to 0.8 units lower than in distilled water. Measuring soil pH in these salt solutions has the added advantage of maintaining flocculation which minimizes errors caused by liquid junction potentials

D. Equipment

1. pH meter with suitable electrode
2. Paper cups

LIME REQUIREMENT SMP Buffer Method

A. Reagents

1. SMP buffer solution - Using a 1-L volumetric flask, completely dissolve 1.8 g of ground para-nitrophenol in 500 mL distilled water. Add 2.5 mL or 2.8 g of triethanolamine (weigh rather than pipette this viscous liquid). Then dissolve 3.0 g potassium chromate (K_2CrO_4), 2.0 g calcium acetate ($\text{Ca}(\text{OAc})_2 \cdot \text{H}_2\text{O}$) and 53.1 g calcium chloride dihydrate ($\text{CaCl}_2 \cdot 2\text{H}_2\text{O}$) in the solution. Bring to 975 mL volume with distilled water and stir overnight with magnetic stirrer. Adjust the solution to pH 7.5 with 0.1 N NaOH if necessary. Bring to 1 L volume with distilled water. This solution is usually made in 8 L quantities for convenience.

CAUTION: Triethanolamine and potassium chromate can be hazardous. Read label before use.

2. Sodium hydroxide, 0.1 N NaOH - Dissolve 4.0 g of NaOH pellets in about 500 mL distilled water. Allow to cool to room temperature and bring to 1 L volume.
3. Hydrochloric acid, 0.1 N HCl - Dilute 8.3 mL of concentrated HCl to 1 L volume with distilled water.
4. Phosphate buffer, pH 6.860 - See pH.

B. Procedure

1. Weigh 5.0 g of soil into paper cup or beaker. Generally samples are placed in rows of six to accommodate continuous stirring and reading of samples.
2. Add 5.0 mL of distilled water. Stir (leaving a stir rod in each sample) and allow to soak for 30 min.
3. Standardize the pH meter, described in B.4 of pH Procedure.
4. Add 10 mL of SMP buffer solution and stir every 5 min during the ensuing 20 minute period.
5. Immediately following the final stirring (20 min after addition of SMP buffer solution), insert the electrodes and observe the pH reading of the suspension, swirl gently and observe the subsequent reading. Continue until pH readings are constant, then record the pH reading to the nearest 0.1 unit.
6. Between readings, thoroughly rinse electrodes with distilled water and pat dry.

C. Comments

Reading the pH of the soil-buffer solution between 20 and 25 min after the addition of the SMP buffer is necessary because the pH of the suspension will continue to decrease over time. The electrodes should be rinsed with 0.1 N HCl and distilled water occasionally when making a series of determinations to eliminate increased pH readings caused by contamination of the electrodes.

The method outlined is a modification of the method described by McLean (1982).

D. Equipment

1. pH meter and suitable electrode
2. Paper cups

EXTRACTABLE PHOSPHORUS Sodium Bicarbonate Method

Note: This method is used for all samples received from east of the Cascade Mountains.

A. Reagents

1. Sodium bicarbonate, 0.5 M NaHCO_3 - Using a 1-L volumetric flask, dissolve 42.01 g NaHCO_3 in 500 mL of distilled water and make up to volume. Cover and store overnight. Adjust the pH to 8.5 with 1 M NaOH. Cover the surface of the solution with an approximately 1 inch thick film of purified mineral (paraffin) oil to seal the solution from the air. When stored in a glass container, prepare a fresh solution monthly. A longer storage period is acceptable when the solution is stored in a polyethylene container. Check the pH of the solution each month, and adjust the pH if necessary. (See Section D, Comments.)
2. Ammonium paramolybdate - In a 1-L flask dissolve 15.0 g $(\text{NH}_4)_6\text{Mo}_7\text{O}_{24}\cdot 4\text{H}_2\text{O}$ in 300 mL of warm distilled water (60C). After cooling, filter the solution if turbidity is evident, adding 342 mL of concentrated HCl gradually

while swirling; bring to volume. This solution contains enough concentrated HCl so that a 2 mL aliquot of ammonium paramolybdate solution has sufficient acid to neutralize the NaHCO_3 in a 2 mL aliquot of soil extract.

3. Stannous chloride
 - a. Stock solution - Dissolve 10.0 g $\text{SnCl}_2\cdot 2\text{H}_2\text{O}$ in 25 mL of concentrated HCl. Prepare fresh every two months or less. Use large reagent crystals for preparing the solution rather than fine powder, and store the stock solution in a refrigerator.
 - b. Dilute solution - Add 0.5 mL aliquot of the stock solution to 66 mL of distilled water. Prepare this solution fresh daily.
4. Standard phosphate solutions
 - a. Standard stock solution (50 ppm P) - Dissolve 0.2195g oven dried KH_2PO_4 in 500 mL distilled water and dilute to 1 L volume.
 - b. Standard work solutions - Pipette the following aliquots of 50 ppm P stock solution into 100 mL volumetric flasks. Bring to volume with NaHCO_3 extracting solution.

<u>mL stock solution</u>	<u>ppm P work solution</u>
1	0.5
2	1.0
4	2.0
6	3.0
10	5.0

5. Sodium hydroxide, 1 M NaOH - Dissolve 40 g NaOH pellets in 500 mL distilled water and dilute to 1 L volume.

B. Procedure

1. Weigh or scoop 2.0 g of soil into a 50 mL extracting bottle and add 40 mL of NaHCO_3 extracting solution.
2. Shake the sample for 30 min, remove the sample from the shaker immediately after it stops. Decant the contents of the bottle into a filter funnel fitted with a Whatman No. 42 or equivalent filter paper. Refilter the extract if it is not clear.
3. Pipette 2.0 mL of the filtrate into a 25 mL colorimeter tube. Automatic pipettes are suitable for dispensing the small volumes used in all of the following steps of this procedure.
4. Add 2.0 mL of ammonium paramolybdate solution to each tube and mix well using a Vortex mixer. Remove all traces of the molybdate solution from the neck of the flask by washing with 5.0 mL of distilled water. Vortex for 5 s.
5. Add 0.5 mL of the dilute SnCl_2 solution, mix immediately.
6. Read color intensity in the colorimeter¹ set at a wavelength of 660 nm, at least 10 min but not more than 30 min after addition of the SnCl_2 solution.
7. Prepare a calibration curve using steps 3-6, but substitute 2.0 mL aliquots of the 0.5 to 5 ppm P standard solutions for the soil extract. Report the results in ppm P (mg kg^{-1}) in the soil sample.

C. Calculations

ppm P in the soil sample = ppm P in the soil extract x 20

D. Comments

This method for extractable P follows a procedure outlined by Olsen and Sommers (1982) with the following exceptions:

1. The ammonium paramolybdate solution contains sufficient HCl to neutralize the NaHCO₃ 2 mL aliquot of extractant. This eliminates the step of acidifying the aliquot with H₂SO₄.
2. A colorimeter tube is used for the color development step rather than a volumetric flask.
3. Stannous chloride is used as the reducing agent instead of ascorbic acid.

When P is extracted from soil with a 0.5 M NaHCO₃ solution at an approximate pH of 8.5, the concentrations of calcium (Ca), aluminum (Al), and iron (Fe) in solution are maintained at low levels. A decrease in activity or concentration of soluble Ca, Al, and Fe allows extraction of more soluble phosphate.

An increase in shaker speed or temperature of the extractant may cause an increase in P extracted from the sample. Normally, for routine testing, the extraction is performed at room temperature, though it may vary seasonally. The OSUSTL uses a constant-speed reciprocating shaker, which has a 2-inch stroke and operates at 200 oscillations per minute.

When exposed to the atmosphere, NaHCO₃-extracting solution increases over time. When pH of the extractant exceeds 8.5, an increase in extractable soil P is anticipated. Spreading a layer of mineral oil spread over the surface of the extracting solution will decrease the rate pH will change. Prolonged storage of the NaHCO₃ extractant in glass may also allow a pH increase. When glass storage vessels are used, check the pH of the solution at least monthly; if pH of the solution exceeds 8.5, prepare a new solution.

E. Equipment

1. Spectrophotometer
2. Flow-through cell or cuvettes
3. Extraction bottles
4. Filtration vials
5. Vortex mixer
6. Reciprocating shaker

EXTRACTABLE PHOSPHORUS Dilute Acid-Fluoride Method (Bray-P1)

Note: This method is used for all samples received from west of the Cascade Mountains, including Hood River County.

A. Reagents

1. Ammonium fluoride, 1 N NH₄F - Dissolve 74 g of NH₄F in distilled water and dilute the solution to 2 L. Store the solution in a polyethylene bottle.
2. Hydrochloric acid, 0.5N HCl - Dilute 103 mL of concentrated HCl to a volume of 2500 mL with distilled water.
3. Extracting solution - Add 1350 mL of 1.0 N NH₄F and 2250 mL of 0.5 N HCl to 45 L of distilled water. This produces

a solution of 0.03 N NH₄F and 0.025 N HCL. It will keep indefinitely.

4. Standard phosphate solutions

- a. Standard stock solution, 100 ppm P - Dissolve 0.4393 g of oven dry KH₂PO₄ in 500 mL of distilled water and dilute to a volume of 1 L.
- b. Standard work solution - Pipette the following aliquots of 100 ppm stock solution into 100 mL volumetric flasks. Bring to volume with NH₄F extracting solution.

Aliquot mL	ppm P of solution
5	5
10	10
15	15
20	20

B. Procedure

(i) Automated Colorimetric Analysis (OSU Procedure)

1. Weigh 2.9 g (or scoop 2 g) of soil into a 50 mL extracting bottle and add 20 mL of the extracting solution.
2. Shake for 60 sec. and filter immediately using Whatman No. 42 or equivalent filter paper.
3. The concentration of P in the extract solution is determined on a ALPKEM rapid flow analyzer No. RFA-300 which relies on molybdate and antimony in acid to form a complex with ortho phosphate to yield a blue color.

(ii) Manual Colorimetric Analysis

1. Use same procedure as for sodium bicarbonate method.

C. Calculations

$$\text{ppm P in soil sample} = \text{ppm P in soil extract} \times 7$$

D. Comments

The dilute acid-fluoride method for P follows a method described by Olsen and Sommer (1982). OSUSTL modifications are a 2.9 g weight used with a 60 second shaking time.

The dilute acid-fluoride extractant tends to dissolve Al and Fe phosphates in soil. The dissolution of Al and Fe phosphates occurs very rapidly and probably results from the fluoride anion complexing these metal cations in the acid solution. Interference in the development of the color complex occurs if appreciable amounts of Al, Fe (excess of 100 ppm), and molybdate are present. The fluoride ion may also interfere with color development when present in excess of 50 ppm. To minimize interferences, standards are made using the extracting solution.

E. Equipment

1. Auto-analyzer or spectrophotometer
2. Reciprocating shaker
3. Filtration vials
4. Extraction bottles

**EXTRACTABLE CALCIUM, MAGNESIUM,
POTASSIUM, AND SODIUM
Ammonium Acetate Method**

A. Reagents

1. Ammonium acetate extracting solution, neutral, 1 N - Commercial ammonium acetate is purchased for ease of handling and to reduce ammonia contamination in the lab. To mix add 77.1 g ammonium acetate per liter of solution, usually mixed in 45 L quantities. This solution does not have to be neutralized as it does when acetic acid and ammonium hydroxide are used.
2. Lithium - lanthanum chloride solution (reagent grade $\text{LaCl}_3 \cdot 7\text{H}_2\text{O}$ and LiCl), dissolve 200 g $\text{LaCl}_3 \cdot 7\text{H}_2\text{O}$ and 50 g LiCl in a 22 L container with 5 L distilled water. Fill to the 22-L mark and mix.
3. Standard solutions
 - a. Standard stock solutions. These can be prepared from commercial standard solutions which are available through most chemical suppliers, or can be prepared as follows:
 - (i) Calcium (500 ppm Ca) - Dissolve 1.249 g of CaCO_3 in 1:1 HCl and evaporate to dryness on a hot plate. Dissolve the residue and bring to exactly 1 L with distilled water.
 - (ii) Magnesium (500 ppm Mg) - Dissolve 0.50 g pure Mg ribbon in 1:1 HCl and evaporate to dryness on a hot plate. Dissolve the residue and then dilute to 1L with distilled water.
 - (iii) Potassium (500 ppm K) - Prepare a standard solution of K by dissolving 0.9535 g oven dried KCl in a small volume of distilled water and diluting to 1 L with distilled water.
 - (iv) Sodium (500 ppm Na) - Prepare a standard solution of Na by dissolving 1.271 g NaCl in a small volume of distilled water and diluting to 1 L with distilled water.
 - b. Standard work solutions⁴ K, Ca, Mg, and Na - Pipette the following aliquots of 500 ppm stock solutions into 100 mL volumetric flasks.

Dilutions of stock solutions for standard preparation.

Flask or Standard No.	Ca		Mg	
	Aliquot mL	ppm in solution	Aliquot mL	ppm in solution
1	5	25	1.0	5.0
2	15	75	1.5	7.5
3	25	125	2.0	10.0
4	35	175	2.5	12.5
5	70	350	7.5	37.5

Flask or Standard No.	Na		K	
	Aliquot mL	ppm in solution	Aliquot mL	ppm in solution
1	1	5	2	10
2	2	10	3	15
3	4	20	4	20
4	5	25	6	30
5	10	50	12	60

Bring to 100 mL volume with ammonium acetate. Mix thoroughly and store in plastic bottles.

B. Procedure

1. Weigh or scoop 2.0 g of soil into a 50-mL extracting vessel. Add 40 mL of the ammonium acetate extracting solution and place the extracting vessel containing the sample on the shaker for 30 min.
2. Filter through a Whatman No. 40 or equivalent filter paper.
3. K, Ca, Mg and Na. Using a Custom Lab Equipment diluter dispenser or the equivalent, dilute a 0.5 mL aliquot of the sample filtrate with 12 mL of LaCl_3 -LiCl solution (a 25-fold dilution). Prepare standards by substituting 0.5 mL of standard K, Na, Ca or Mg work solutions for the sample filtrate. The blank is made by diluting the ammonium acetate extracting solution.
4. Calibrate the atomic absorption spectrophotometer⁵ with the standard work solutions according to instrument instructions.
5. Report Ca, Mg, K and Na in millequivalents per 100 g, ppm or mg/kg of soil.

C. Calculations

ppm in the soil sample = ppm in the soil extract solution x 20

mcq per 100g of sample = ppm in the soil sample divided by equivalent weight (K=390, Ca=200, Mg=120, Na=230)

D. Comments

The procedure for determining extractable cations with neutral 1 N ammonium acetate is a modification of the procedure outlined by Knudsen et al. (1982) for exchangeable K. The modification is the equilibration of a sample with one extracting solution (1:20 ratio of soil to extractant) rather than three different extractions, as specified in the original procedure. A further modification is the dilution of the soil extract with a joint lanthanum chloride and lithium chloride solution.

The single extraction technique for cations in non-calcareous soil results in values which are equivalent to at least 95% of the values obtained by the process of multiple extraction. For samples which contain carbonates of Ca or Mg, the multiple extraction with ammonium acetate may dissolve these carbonates and result in higher values for Ca and Mg than are obtained with a single extraction. However, for purposes of routine soil testing, there is usually no interest in determining the extractable Ca and Mg in alkaline samples which contain free lime.

Interferences caused by refractory compound formation and ionization are minimized by the dilution of the soil extract with lanthanum chloride and lithium chloride, respectively. The addition of lanthanum chloride minimizes the formation of Ca and Mg refractory compounds. Lithium chloride is added for Na and K determinations to minimize ionization interferences. In the past, these have been two separate solutions but it was determined that they could be mixed without sacrificing analytical accuracy. For some samples, the use of this mixture tends to stabilize readings and improve precision.

E. Equipment

1. Atomic absorption instrument
2. Filtration vials
3. Extraction bottles
4. Reciprocating shaker
5. Diluter-dispenser

HOT-WATER EXTRACTABLE BORON Azomethine H Method

A. Reagents

1. Buffer masking agent - Completely dissolve 250 g ammonium acetate (reagent grade $\text{NH}_4\text{C}_2\text{H}_3\text{O}_2$), 25 g tetrasodium salt of ethylene-dinitrilotetraacetic acid (Na_4 -EDTA), and 10 g disodium salt of nitrilotriacetic acid (Na_2 -NTA) in 400 mL distilled water in a 1-L beaker using a magnetic stirrer. Add 125 mL glacial acetic acid very slowly, while stirring. The temporary acidic conditions may cause a slight precipitation of the EDTA salts. Continue to stir the solution until all the EDTA redissolves. Do not heat the solution. Adjust the buffer to a pH of 5.4 to 5.6 with acetic acid or ammonium hydroxide as necessary. If the spectrophotometer is equipped with an aspirating flow-cell, add six drops of Brij-35 surfactant (ALPKEM) to 250 mL buffer masking agent. Prepare this solution every two months.
2. Azomethine-H solution - Dissolve 0.9 g azomethine-H reagent (Pierce Chemical Co., Rockford, IL) and 2.0 g ascorbic acid ($\text{C}_6\text{H}_8\text{O}_6$) in about 50 mL of distilled water. A hot tap water bath facilitates dissolution. Bring to 100 mL volume with distilled water. Prepare this solution fresh daily.

Note: Azomethine-H reagent may also be prepared in the laboratory.

3. Calcium chloride extracting solution, 0.02 M - Dissolve 2.84 g calcium chloride dihydrate ($\text{CaCl}_2 \cdot 2\text{H}_2\text{O}$) in about 700 mL distilled water, then bring to one liter volume. Store in plastic container.
4. Boron standard solutions - All standard solutions should be stored in plastic bottles.
 - a. Standard solution I, 500 ppm B - Pipette 5.0 mL of 5000 ppm aqueous boron standard solution (available commercially) into a 50 mL volumetric flask. Bring to volume with distilled water. A 500 ppm B standard solution can also be prepared by dissolving 0.8820 g oven-dry re-crystallized sodium tetraborate (reagent $\text{Na}_2\text{B}_4\text{O}_7 \cdot 10\text{H}_2\text{O}$) in distilled water and diluting to 200 mL.
 - b. Standard solution II, 5 ppm B - Pipette 5.0 mL of standard solution I (500 ppm B) into a 500 mL volumetric flask. Bring to volume with distilled water.
 - c. Standard work solution - Prepare work solutions by pipetting the following aliquots of standard solution II (5 ppm B) into 100 mL volumetric flasks. Bring to volume with CaCl_2 extracting solution.

<u>mLs Stock II (5 ppm B)</u>	<u>Standard Work Solution (ppm B)</u>
4	0.20
8	0.40
12	0.60
20	1.00
28	1.40
40	2.00

B. Procedure

1. Weigh or scoop 15 g of soil into a sealable plastic bag (heat sealed boilable bags or ziplock freezer bags work). Add 30 mL of CaCl_2 extracting solution.
2. Place plastic bags into boiling water and leave for 10 min. The OSUSTL uses a porcelain canning pot with cover.
3. Remove plastic bags, let cool to room temperature and filter the contents through a Whatman No. 42 or equivalent filter paper.
4. Pipette 4.0 mL of soil extract into a 12 mL polyethylene sample vial.
5. Add 1 mL of buffer masking agent and vortex.
6. Add 1 mL of azomethine-H solution and vortex. Allow color to develop for at least 1 hour but no longer than 3 hours.
7. Prepare standard curve following steps 4-6, substituting 4.0 mL of standard work solution for soil extract. A blank is prepared in the same manner using 4.0 mL CaCl_2 extracting solution instead of the soil extract.
8. For samples with a yellow extract: Prepare a second sample solution and blank following steps 4 and 5. Add 1.0 mL of distilled water in place of azomethine-H solution and vortex well. The blank for this determination consists of 5.0 mL CaCl_2 extracting solution and 1.0 mL buffer masking agent.
9. Read all color intensities on a spectrophotometer set at 420 nm. Read immediately after vortexing.

C. Calculations

ppm B in soil = (ppm B extract - ppm B in yellow extract) x 2

D. Comments

A method described by Bingham (1982) is used here with adaptation to the use of plastic bags as described by Mahler et al. (1983). It was determined that plastic bags are more suitable and less expensive than boron free glassware, which is no longer obtainable. The pH of the buffer was originally prescribed as 5.2, but 5.4 to 5.6 is adequate. Further reductions in pH only increases the difficulty of keeping the EDTA in solution.

The EDTA and NTA chelates eliminate interferences from Al, Fe, and Cu. The concentration of these chelates should be effective for levels of these elements commonly found in soil extracts.

The azomethine-H should be added quickly so that time for color development is equal for all tubes. A constant check must be maintained on linearity and drift of the standard curve when analyzing a large batch of samples. Correction for a yellow extract as described here is probably legitimate for only

a mild yellow color and is insufficient for some of the deep brown or yellow extracts occasionally obtained. For these ICP analysis is preferable. Acid washing of all glassware is recommended to minimize the potentials for boron contamination.

D. Equipment

1. Spectrophotometer
2. Flow through cell or cuvettes
3. Filtration vials
4. Hot plate and boiling container with cover
5. Vortex stirrer

ORGANIC MATTER Walkley-Black Method

A. Reagents

1. Potassium dichromate, 1 N $K_2Cr_2O_7$ - Dissolve 49.04 g of reagent grade $K_2Cr_2O_7$ in 500 mL distilled water and dilute the solution to a volume of 1 L.
2. Ferrous ammonium sulfate, 0.4 N $Fe(NH_4)_2(SO_4)_2 \cdot 6H_2O$ - Dilute 40 mL concentrated H_2SO_4 in 500 mL distilled water. Dissolve 159.6 g $Fe(NH_4)_2(SO_4)_2 \cdot 6H_2O$ in the acid solution; cool the solution and dilute it to a volume of 1 L. Determine the normality periodically by titrating against the $K_2Cr_2O_7$ solution. Store in opaque bottle as light affects this solution.
3. O-phenanthroline ferrous sulfate complex indicator, 0.025 M - This solution is also referred to as 1,10 phenanthroline iron (II) sulfate and is commercially available under the trade name "Ferroin."
4. Phosphoric acid, 85 percent, H_3PO_4 .
5. Sulfuric acid, concentrated, not less than 96 percent H_2SO_4 .

B. Procedure

1. Pass the soil sample through a 0.5 mm sieve and weigh out 0.50 g of soil into a 500-mL Erlenmeyer flask.
2. Add 10 mL of 1 N $K_2Cr_2O_7$ solution and swirl the flask to gently disperse the soil in the solution. Take care not to throw sample onto sides of flask.
3. Rapidly add 20 mL of concentrated H_2SO_4 . Swirl for 10 seconds. Let cool uniformly to room temperature, at least 20 min.
4. Dilute to approximately 150 mL with distilled water and add 10 mL of concentrated H_3PO_4 . The addition of H_3PO_4 is optional and the OSUSTL omits this step for routine analysis.
5. Add 6 drops of O-phenanthroline indicator to the solution. Titrate with the ferrous ammonium sulfate solution (FAS) until the color changes from yellow or yellow-green to blue to finally a reddish brown endpoint. Record the volume (mLs) of FAS used to reach the endpoint.
6. Analyze a blank simultaneously following steps 2-5.

C. Calculation

Calculate the percent organic matter as follows:

$$\%OM = (\text{Blank-reading}) \times \frac{13.6}{\text{blank}}$$

Calculate the percent organic carbon as follows:

$$\%OC = \%OM \times 0.58$$

D. Comments

The wet oxidation method for determining organic matter in soil is the same method as described by Nelson and Sommers (1982). The only modification involves the use of the O-phenanthroline in place of the diphenylamine indicator.

Grinding of the soil sample to pass a 0.5 mm sieve facilitates obtaining a representative subsample, increasing surface area and ridding the sample of ground plant material. If more than 75% to 80% of the total dichromate reagent is reduced by the oxidizable material in the sample, the entire analysis must be repeated using a smaller soil sample.

The soil is digested with the dichromate and sulfuric acid mixture by the heat of dilution. For precise results the sulfuric acid should be added rapidly and the flasks should be cooled uniformly. Once these steps are accomplished, variations in reaction time from 20 to 40 min do not appreciably affect the results.

For soils or other materials high in organic matter, the organic matter content may be more accurately determined using the Ignition method presented in the Appendix.

E. Equipment

1. Titration apparatus
2. Lighted stirring plate

SOLUBLE SALTS
Electrical Conductivity Method

A. Reagent

1. Potassium chloride reference solution, 0.01 N KCl - Dissolve 0.7456 g of KCl in distilled water and dilute the solution to a volume of 1 L at 25 C. This solution has a conductivity of 1.4118 mmhos per cm (ds/m).

B. Procedure

1. Place 30 to 50 mL of soil in a 10 oz paper cup; add distilled water while stirring to prepare a saturated soil paste. (At saturation, the soil paste glistens as it reflects light and it flows slightly when the container is tipped. The paste slides freely and cleanly off the spatula unless the soil has a high clay content.)
2. Allow the saturated soil to stand at least 30 min. Then ascertain that the above criteria for saturation are still evident. Free water should not collect on the soil surface, nor should the paste stiffen markedly or lose its glisten. Remix the sample, if necessary, by adding either additional water or soil to obtain a saturated paste.
3. Transfer the saturated soil paste to a Buchner funnel fitted with a Whatman No. 42 filter. By vacuum filtration⁶, collect an aliquot of the saturation extract in a 25 mL receiving flask.
4. Using the reference solution, calibrate the conductivity meter⁷ according to instrument instructions.
5. Record the electrical conductivity (EC) reading for the saturation extract when it has reached the same temperature as the reference solution.

C. Comments

The procedure for determining total soluble salts follows closely a method described by Rhoades (1982b). For an appraisal of soil salinity, the extraction can usually be made a few minutes after the saturated paste is prepared. The recommended time lapse between preparation of the soil paste and extraction is several hours for gypsiferous samples and from 4 to 16 hr in all cases where the chemical constituents are to be determined in the extract. Determination of chemical constituents in the extract requires a larger soil sample (200-400 g soil) than for soluble salts alone. If the initial filtrate is turbid, it can be discarded or refiltered through a clean sheet of filter paper.

The Solu-Bridge used in the OSUSTL is designed specifically for determining the conductivity of saturation extracts. When the compensator dial is set on the temperature of the solution, the conductivity dial at balance indicates directly the electrical conductivity at 25 C. A calculation to obtain the result is unnecessary.

E. Equipment

1. Conductivity meter
2. Suction filtration apparatus

CATION EXCHANGE CAPACITY (CEC)
Ammonium Acetate Method

A. Reagents

1. Ammonium acetate extracting solution, neutral, 1 N - Prepare according to the specifications outlined in the ammonium acetate method for extractable cations.
2. Ethanol, 95%
3. Hydrochloric acid, 0.1 N HCl - Dilute 8.3 mL of concentrated HCl reagent to 1 L with distilled water.

B. Procedure

1. Weigh 10 g of soil into a 125 mL Erlenmeyer flask; add 50 mL of ammonium acetate solution and place the flask containing the sample on the shaker for 30 min.
2. Connect a 1-L vacuum extraction flask to a Buchner funnel fitted with a Whatman No. 5 or equivalent filter paper. Moisten the filter paper with distilled water.
3. Transfer the soil suspension into the Buchner funnel and leach the sample with 175 mL of 1 N ammonium acetate. This soil extract may be analyzed for extractable K, Ca, Mg, and Na.
4. Rinse the excess ammonium acetate from the soil sample in the Buchner funnel by leaching with a total volume of ethanol and discard the leachate. Note: Be sure to gently fill funnel to remove all excess ammonium and allow it to drain until only damp soil remains. Continue adding alcohol in this manner until 200 mL of ethanol has been used.
5. Change to a clean 500-mL suction flask and leach the soil sample with 225 mL of 0.1 N HCl to replace the exchangeable ammonium. Bring leachate to volume in a 250 mL volumetric flask using distilled water.
6. The concentration of ammonium-N in the final leachate is determined with an ALPKEM rapid flow analyzer (RF-300), which relies on ammonium to complex with salicylate to form indophenol blue (Technicon Method No. 334-74A/A). This color is intensified with sodium nitroprusside and measured at 660 nm. This determination can also be made using the Kjeldahl distillation method (see Appendix).

C. Calculation

$$\text{CEC in meq per 100 g of soil} = \frac{\text{ppm NH}_4\text{-N in leachate}}{14} \times \frac{0.25}{14} \times \frac{100}{\text{sample size (g)}}$$

ppm NH₄-N in leachate is determined using a standard curve.

D. Comments

The procedure used is essentially the same as that of Scholtenberger (1945) except that determination of NH₄-N is done spectrophotometrically rather than by Kjeldahl distillation and titration. To determine the NH₄-N content using the Kjeldahl distillation method, follow steps 1-5 above, then proceed to Appendix. Care must be taken not to allow soil to dry and

crack between alcohol leachings, as this could result in incomplete removal of excess $\text{NH}_4\text{-N}$. A similar procedure is described by Rhoades (1982a).

E. Equipment

1. Buchner funnels and source of vacuum
2. Auto analyzer or Kjeldahl distillation equipment
3. Vacuum flasks

TOTAL NITROGEN (TN) Kjeldahl Method

A. Reagents

1. Sulfuric acid, concentrated H_2SO_4 - reagent grade
2. Digestion catalyst - Mix together 1000 g of ground sodium sulfate (reagent anhydrous Na_2SO_4) or potassium sulfate, 25 g cupric sulfate (reagent anhydrous CuSO_4), and 10 g of reagent selenium (Se) powder. Packets of prepared catalyst can be purchased.

CAUTION: DO NOT BREATHE CuSO_4 and Se dust..

B. Procedure

1. Weigh 3.0 g of soil into a 75 mL volumetric digestion tube. Use 1.0 g of soil if sample is greater than approximately 20% in organic matter.
2. Add a 3 g scoop of digestion catalyst and mix thoroughly with the dry soil.
3. Add 10 mL of concentrated H_2SO_4 to the soil-catalyst mixture. *Note: It is essential that all dry material be completely moistened and well mixed with the acid to insure complete digestion.*
4. Prepare a blank with each set of samples analyzed by following steps 2-3 above using no soil. Allow the samples and blank to stand overnight.
5. Place tubes on a digestion block⁸ at 150 C. Check samples every 20 min for foaming. After one hour (or more if foaming persists), raise temperature to 250 C, and continue digestion for one hour. After one hour at 250 C raise temperature to 350 C and heat until samples are completely digested, usually about two additional hours. At completion, mineral soils will be greyish-white while organic soils will be blue-green in color.
6. Remove samples from block and leave under a fume hood until cool. Then add 10-20 mL distilled water to each tube to keep samples from hardening.
7. The ammonium-N content of the digest solution is determined with an ALPKEM rapid flow analyzer (RF-300) which relies on ammonium to complex with salicylate to form indophenol blue (Technicon Method No.334-74A/A). This color is intensified with sodium nitroprusside and measured at 660 nm. This determination can also be made using the Kjeldahl distillation method (see Appendix). For samples to be analyzed on an auto analyzer, continue with steps 8-9 and determine total N using calculation in Part C.

8. Bring samples to volume with deionized water in 75 mL digestion tubes and mix.
9. Obtain a clear digest solution for analysis either by allowing samples to settle overnight and pipetting an aliquot or by filtering through an acid washed filtering apparatus fitted with Whatman No. 042 or equivalent filter paper. Digest solutions may be refrigerated prior to analysis.

C. Calculation

$$\% \text{ Total Nitrogen} = (\text{ppm } \text{NH}_4^+ \text{-N in digest solution}) \times \frac{75 \text{ mL}}{\text{sample size (g)}} \times \frac{1}{10,000}$$

D. Comments

The Kjeldahl method outlined by Bremner and Mulvaney (1982) is modified by eliminating the water from the digestion step. One further modification is the determination of $\text{NH}_4\text{-N}$ spectrophotometrically rather than by Kjeldahl distillation and titration. To determine the $\text{NH}_4\text{-N}$ concentration using the Kjeldahl distillation method, follow steps 1-6 and then proceed to Appendix.

E. Equipment

1. Digestion block
2. Digestion tubes
3. Autoanalyzer or Kjeldahl distillation unit

AMMONIUM AND NITRATE NITROGEN KCl Extraction Method

A. Reagents

1. Potassium chloride extracting solution, approximately 2 N KCl - Dissolve 150 g of reagent KCl in 500 mL distilled water and dilute to a volume of 1 L.

B. Procedure

1. Place 20 g of soil into a 250 mL extracting bottle and add 75 mL of 2 N KCl extracting solution. *Note: If using the Kjeldahl distillation method, add 150 mL of extracting solution.* Shake the vessel on a mechanical shaker for one hour. Remove from shaker and allow the soil-KCl suspension to settle (about 30 min).
2. Filter the extract solution through Whatman No. 42 or equivalent filter paper. To minimize contamination by filter paper, it is first leached with 20-50 mL of KCl solution. If the extract cannot be analyzed on the same day as prepared, store in a refrigerator or freezer until analysis can be performed.
3. The ammonium-N content of the extract is determined with an ALPKEM rapid flow analyzer (RF-300) which relies on ammonium to complex with salicylate to form indophenol blue (Technicon Method No. 334-74A/A). This color is intensified with sodium nitroprusside measured at 660 nm. This determination can also be made using the Kjeldahl distillation method (see Appendix).

4. The nitrate-N content of the extract is determined with an ALPKEM rapid flow analyzer (RF-300) which reduces nitrate to nitrite via a cadmium reactor then complexes nitrite with sulfanilamide and N-(1-Naphthyl)-ethylenediamine dihydrochloride to form a red-purple color that is measured at 540 nm (Technicon Method No. 329-74W/A). This determination can also be made using the Kjeldahl distillation method (see Appendix).

C. Calculation

$$\text{ppm NH}_4\text{-N or NO}_3\text{-N in soil sample} = (\text{ppm NH}_4\text{-N or NO}_3\text{-N in filtrate} \times 3.75)$$

D. Comments

The method outlined by Keeney and Nelson (1982) for determining ammonium and nitrate-N is used with a modification in which 75 mL of KCl and 20 g of soil are used instead of 100 mL and 10 g soil. To determine NH₄-N or NO₃-N concentration using the Kjeldahl method, follow steps 1-2 and then proceed to Appendix.

The extended period of shaking the soil sample with 2 N KCl according to the specifications of Bremner's original procedure permits the simultaneous extraction of ammonium and nitrate.

E. Equipment

1. Autoanalyzer or Kjeldahl distillation apparatus
2. Reciprocating shaker
3. Filtration Vials
4. Extraction Bottles

EXTRACTABLE ZINC, COPPER, AND MANGANESE DTPA Method

A. Reagents

1. Diethylenetriaminepentaacetic acid, 0.025 M DTPA - Mix 9.83 g DTPA in glass-distilled water and dilute to a volume of 1 L.
2. Triethanolamine, 0.5 M TEA - Mix 74.60 g TEA in glass-distilled water and dilute to a volume of 1 L.
3. Calcium chloride, 0.05 M CaCl₂ - Dissolve 5.55 g anhydrous CaCl₂ in glass-distilled water and dilute to 1 L.
4. DTPA extracting solution, 0.05 M DTPA, 0.1 M TEA, and 0.01 M CaCl₂ - Combine reagents from steps 1, 2, and 3, and dilute to 5 L with glass-distilled water. Adjust the resulting solution after it has set for 12 hr to pH 7.3 with concentrated HCl. Two mL of concentrated HCl is needed to change the pH of the DTPA solution 0.1 units. Store the solution in the refrigerator.
5. Standard solutions
 - a. Standard stock solutions - These are easily made from commercial standard solutions which are available through most chemical suppliers, or can be prepared as follows:
 - (i) Zinc (100 ppm Zn) - Weigh 0.1000 g of pure Zn metal

(30-mesh, analytical reagent) into a 1-L volumetric flask. Add 50 mL of Zn-free water and 1 mL of concentrated H₂SO₄. When the Zn has dissolved, make to volume with DTPA extracting solution.

(ii) Copper (100 ppm Cu) - Dissolve exactly 0.1000 g of pure metallic Cu in 15 mL of 3 N HNO₃ at room temperature in a covered 125-mL Erlenmeyer flask. When the solution has cooled, add 1 mL of concentrated H₂SO₄ and evaporate the solution cautiously until SO₄ fumes are evolved. Cool the solution again; dilute it cautiously with 10 to 15 mL of glass distilled water and again evaporate until it fumes SO₄. Finally, when the solution has cooled, dilute it cautiously with water, transfer it quantitatively to a 1-L flask and dilute the solution to volume with DTPA extracting solution.

(iii) Manganese (100 ppm Mn) - Dissolve 0.2880 g of dry, pure KMnO₄ in about 250 mL of H₂O in a 1-L beaker. Add 20 mL of 18 N H₂SO₄; heat the solution to boiling. Add solid Na₂SO₄ until the color of permanganate disappears (avoid a large excess of Na₂SO₄) and boil off the SO₂. Cool the solution, transfer to a 1-L volumetric flask, and bring to volume with DTPA extracting solution.

b. Standard work solutions - Prepare standard work solutions by pipetting the following amounts of 100 ppm standard stock solutions into 100 mL volumetric flasks and diluting to volume with DTPA extracting solution:

Dilutions of stock solutions for metal standard preparation.

Zn		Cu		Mn	
mL 100 ppm Zn	ppm Zn in solution	mL 100 ppm Cu	ppm Cu in solution	mL 100 ppm Mn	ppm Mn in solution
0.5	0.50	1.0	1.00	1.0	1.00
1.0	1.00	2.0	2.00	3.0	3.00
3.0	3.00	5.0	5.00	9.0	9.00

B. Procedure

1. Weigh 10 g of soil into a 125 mL Erlenmeyer flask.
2. Add 20 mL of DTPA extracting solution.
3. Shake on mechanical shaker for two hours at a speed fast enough to keep soil in suspension.
4. Immediately filter through a Whatman No. 42 or equivalent filter paper. Refilter if filtrate is cloudy.
5. Calibrate the atomic absorption spectrophotometer in accordance with instrument instructions using the prepared standard work solutions. The blank is DTPA extracting solution.
6. Determine the concentration of Zn, Cu, and Mn in the filtrate and report as ppm metal in the soil on a dry weight basis.

C. Calculations

$$\text{ppm Zn in soil sample} = \text{ppm Zn in soil extract} \times 2$$

D. Comments

The following precautions are essential to avoid problems of contamination in conducting analyses: (1) All solutions should be prepared with glass-distilled water; (2) All glassware is rinsed with .5 N HCl and then rinsed with glass-distilled water; (3) The filter paper should be checked continuously for presence of zinc, copper, and manganese by analyzing a blank that has been filtered.

The DTPA soil test was developed to measure the availability of zinc, copper, manganese, and iron for plant uptake (Lindsay and Norvell, 1978). Since there have been few reported iron deficiencies in Oregon, the OSU soil testing lab does not routinely measure this nutrient in the extract.

E. Equipment

1. Filtration vials
2. Extraction bottles
3. Reciprocating shaker
4. Atomic absorption spectrophotometer

SULFATE SULFUR (SO₄-S) Ion Chromatograph Method

A. Reagents

1. Standard sulfate-S solutions
 - a. Standard stock solution, 100 ppm SO₄-S - Dissolve 0.5434 g of oven dry potassium sulfate (K₂SO₄) in 500 mL distilled water and dilute to a volume of 1 L.
 - b. Standard working solutions - Prepare work solutions by pipetting the following aliquots of 100 ppm SO₄-S stock solution into 100 mL volumetric flasks. Bring to volume with calcium phosphate extracting solution. The standards are adjusted to suspected concentration of the samples being analyzed. For example, if a sample has a concentration of 3 ppm (.3 ppm in extract) then a standard curve may be developed at .1, .3, .7, and 2 ppm SO₄.

<u>mL 100 ppm stock solution</u>	<u>ppm SO₄-S in work solution</u>
1	1
3	3
7	7
10	10
20	20

2. Calcium phosphate extracting solution, 500 ppm PO₄ - Dissolve 2.17 g calcium phosphate (Ca(H₂PO₄)₂) in 500 mL distilled water and dilute to 1 L volume.

B. Procedure

1. Extraction of SO₄-S
 - a. Weigh 5 g of soil into a 100 mL glass or plastic bottle.
 - b. Add 50 mL of extracting solution and shake vigorously enough to keep soil suspended for 1 hr.
 - c. Filter through Whatman No. 42 filter paper (or equivalent).
2. Determination of SO₄-S Inject 50 uL of extract into ion chromatograph (dionex 2000i) equipped with AS4A anion

exchange column with flow rate set at 2 mL per min. The sulfate peak elutes between 6 and 8 minutes.

C. Calculations

Peak height is integrated by computer and compared to known standards to yield concentration of SO₄ in the extraction solution.

Soil concentration in ppm SO₄ is then calculated by multiplying solution concentration by ten.

D. Comments

The use of an ion chromatograph for sulfate analysis has been shown to be comparable to the methylene blue method (Dick and Tabatabai, 1979). The use of an ion chromatograph also yields greater precision and accuracy than other procedures, especially at low concentrations. The methylene blue method, recommended if access to an ion chromatograph is not available, is described in the Appendix.

EXCHANGEABLE SODIUM Ammonium Acetate Displacement Method⁹

A. Reagents

1. Ammonium acetate extracting solution, neutral, 1 N - Use the same solution prepared for determining ammonium acetate extractable cations.
2. Standard solution, 500 ppm sodium (Na) - Use the same solution which was prepared for determining ammonium acetate extractable Na in the extractable cations section.

B. Procedure

1. Weigh 5 g of soil into a 50-mL plastic centrifuge tube.
2. Add 10 mL of distilled water.
3. Shake by hand three or four times during a 5 to 10-min period to mix.
4. Centrifuge to clarify. Decant supernatant liquid into a paper cup. Test conductivity of supernatant liquid. If over 1.1 mmhos/cm, add 10 mL of distilled water and repeat dilutions until conductivity is below 1.1.
5. Using a stainless steel spatula to loosen the soil in the tube, quantitatively transfer the soil into a 125-mL Erlenmeyer flask using exactly 100 mL of ammonium acetate extracting solution.
6. Swirl every five minutes during a half-hour period.
7. Filter through a Whatman No. 40 or equivalent filter paper.
8. Determine the concentration of Na in the soil extract by the same atomic absorption procedure used to determine ammonium acetate extractable Na.
9. Report the results as exchangeable Na in milliequivalents (meq) per 100 g of soil.

C. Calculations

meq of exchangeable Na per 100 g of soil sample =
ppm of Na in extract x 0.087 (x additional dilution if necessary)

D. Comments

All soil samples should be washed at least once with distilled water to remove any soluble Na. After most of the soluble Na is removed by washing, the conductivity of the wash water should be reduced to approximately 0.9 to 1.1 mmhos/cm (ds/m). The ammonium acetate extractable Na is determined and regarded as an estimate of exchangeable Na. An estimate of exchangeable Na in conjunction with the value for cation exchange capacity serves as a basis for predicting the quantity of soil amendments needed to reclaim sodic soils.

EXCHANGEABLE HYDROGEN Barium Chloride-Triethanolamine Method

A. Reagents

1. Buffer solution, approximately 0.5 N barium chloride ($\text{BaCl}_2 \cdot 2\text{H}_2\text{O}$) and 0.2 N triethanolamine (TEA) - Prepare the following solutions (a and b) and mix together. Protect the buffer solution from CO_2 contamination by storing in a tightly closed plastic container or attaching a tube containing soda lime to the air intake.
 - a. TEA, 0.4 N - Mix 50 mL (56.3 g) of TEA (specific gravity 1.125, about 8N) in 500 mL of distilled water. Partially neutralize the pH to 8.1-8.3 using approximately 150 mL of 1.0 N HCl. Dilute this solution to a volume of 1 L with distilled water.
 - b. BaCl_2 , 1.0 N - Dissolve 125 g $\text{BaCl}_2 \cdot 2\text{H}_2\text{O}$ in 500 mL distilled water and then dilute to a volume of 1 L.
2. Replacement solution, 0.5 N $\text{BaCl}_2 \cdot 2\text{H}_2\text{O}$ in dilute buffer solution - Dissolve 250 g of $\text{BaCl}_2 \cdot 2\text{H}_2\text{O}$ in 2 L of distilled water and dilute to a 4 L volume. Then mix with 20 mL of buffer solution (Reagent 1).
3. Hydrochloric acid, 0.3 N HCl, standardized - Dilute 24.9 mL of reagent concentrated HCl to 1 L with distilled water. Standardize against 0.1000 N sodium carbonate (Na_2CO_3) or 0.1000 N sodium hydroxide (NaOH). These standard base solutions are available through most chemical suppliers, or can be prepared from pure, dry reagent Na_2CO_3 or NaOH. See Appendix for general standardization procedure.
4. Mixed indicator - Dissolve 0.1 g of bromocresol green and 0.02 g of methyl red indicators in 75 mL of 95% ethyl alcohol, then bring to 100 mL volume.

B. Procedure

1. Place at least 10 g of soil in a 125-mL Erlenmeyer flask.
Note: With soils having very high acidity, use 5 g and adjust calculation accordingly.
2. Add 25 mL of buffer solution and swirl the flask occasionally during a 30-minute period to mix the sample suspension.
3. Fit a Buchner funnel which contains a Whatman No. 42 or equivalent paper to a 500-mL vacuum extraction flask. Moisten filter paper with a small amount of buffer solution.
4. Transfer the sample suspension to the Buchner funnel

using an additional 25 mL of buffer solution to completely remove sample from the original 125-mL Erlenmeyer flask. Adjust the filtration rate so that this filtration step requires at least 30 min.

5. When the buffer solution has leached through and only damp soil remains, leach the soil sample with an additional 100 mL of the replacement solution (Reagent 2) by repeatedly adding small increments of the solution to the sample in the funnel.
6. When leaching is completed, remove suction flask and add 10 drops of mixed indicator to the filtrate. Titrate with standardized 0.3 N HCl to a faint pink endpoint. Record the mLs of acid used to reach the endpoint.
7. Titrate a blank solution which contains 50 mL of buffer solution to the same endpoint selected for the sample. The blank determination serves as a reference for the calculation.

C. Calculation

Calculate the result as follows from the volume of standardized HCl used:

Exchangeable hydrogen in meq per 100 g of soil sample =

$$\frac{(\text{mL HCl blank} - \text{mL HCl sample}) \times \text{N HCl} \times 100}{10 (\text{g of sample})}$$

D. Comments

The BaCl_2 -TEA method for determination of exchangeable H as described by Thomas (1982) is followed except for the following modifications:

1. 0.3 N HCl is used instead of 0.2 N HCl.
2. After addition of 25 mL buffer solution into 10 g of soil, the flask is occasionally swirled over a 30 minute period rather than allowing the mixture to stand for 1 hour.
3. Only 25 mL of additional buffer solution is added to remove sample from the original 125-mL Erlenmeyer flask instead of 75 mL of buffer solution.
4. The mixed indicator is slightly different.

This procedure is used as a research tool and is not performed on a routine basis in the OSUSTL.

At the endpoint of the titration, the mixed indicator changes from blue-green through violet and finally to pink. Any stage of the progressive color change may be selected as the endpoint; but the blank and the samples must be titrated to the same endpoint.

The BaCl_2 -TEA extraction estimates the total "potential" acidity which may be related to a potential liming level and a potential CEC. Thomas suggested the use of a KCl extraction method which estimates the neutral and salt-exchangeable acidity. The KCl method is thought to be related to the immediate need for lime and an existing CEC.

E. Equipment

- | | |
|----------------------|------------------------|
| 1. Extraction flasks | 3. Vacuum source |
| 2. Buchner funnels | 4. Titration equipment |

CARBONATE Titrimetric Method

A. Reagents

1. Hydrochloric acid, 2 N HCl - Add 167 mL of concentrated HCl to about 700 mL of distilled water and then dilute to a volume of 1 L.
2. HCl, 1 N - Add 83 mL of concentrated HCl to about 700 mL of distilled water and then dilute to a volume of 1 L.
3. HCl, 0.1 N standardized - Dilute 8.3 mL of concentrated HCl to a volume of 1 L with distilled water. See Appendix for general standardizing instructions.
4. Potassium hydroxide, 2 N KOH - Dissolve 132 g of KOH (85%) in about 700 mL of distilled water and dilute to a volume of 1 L. Protect the solution from atmospheric CO₂ by storing in a tightly stoppered bottle.
5. Bromocresol green indicator - Dissolve 0.1 g of bromocresol green in 100 mL of 95% ethanol.
6. Phenolphthalein indicator - Dissolve 0.05 g of phenolphthalein in 50 mL of ethanol. Add 50 mL of distilled water and mix well.

B. Procedure

1. Weigh 3.0 g of soil into a 250-mL Erlenmeyer flask (or 8 oz French square bottle). If the needle-puncture stopper pops off the glass tube following the addition of the 2 N HCl (Step 4), use 2.0 g of soil. The amount of soil can be further reduced if needed, to as little as 0.5 g. If the stopper pops when using 0.5 g of soil, use the CaCO₃ equivalent procedure used for liming materials, in Appendix.
2. Connect a 5.0 mL beaker to the glass tube below the stopper about 5 mm above the lower end of the tube. Pipette 4.0 mL of 2 M KOH into the 5.0 mL beaker.
3. After stoppering the flask, remove 50 mL of air from the flask via the needle-puncture stopper using a 50-mL gas syringe. Be sure the stopper has been resealed.
4. Inject 20 mL of 2 N HCl into the flask via the needle-puncture stopper with a 20 mL syringe. Be sure stopper has resealed. Swirl the flask gently to mix contents, being careful not to spill the KOH.
5. Allow the flask to stand at room temperature (20-25 C) for 16 to 24 hrs. Then quantitatively transfer the contents of the 5.0 mL beaker into a 125-mL Erlenmeyer flask using 50 mL of distilled water.
6. Add 6 drops of phenolphthalein indicator to the flask and titrate with 1 N HCl until the pink color begins to fade. At this point, titrate with 0.1 N HCl until the solution turns colorless. It is advisable to do one sample at a time, as the pink color of the phenolphthalein tends to fade with time.
7. Add 8 drops of bromocresol green indicator and titrate with the standardized 0.1 N HCl to a pale-yellow endpoint.
8. Determine a blank by following the procedures in the above analysis except do not add soil.

C. Calculations

$$\frac{[(\text{mL HCl}_{\text{sample}} - \text{mL HCl}_{\text{blank}}) \times N \times 0.100]}{\text{wt. of soil sample}} \times 100 = \text{Inorganic carbonate expressed as percent CaCO}_3$$

where mL HCl refers to the amount of acid titrated following the addition of the mixed bromocresol green indicator.

D. Comments

This method follows the same procedure as presented by Bundy and Bremner (1972), except 4 mL of KOH is used instead of 3 mL KOH; N-octyl alcohol is not used and the bromocresol green indicator is made up with ethanol rather than NaOH. These changes should not significantly affect the results.

This procedure determines total carbonate which may be present in compounds such as calcium carbonate, magnesium carbonate and various bicarbonates.

MINERALIZABLE NITROGEN Anaerobic Incubation

A. Reagents

1. Potassium chloride, 2 N KCl - Dissolve 150.0 g of KCl in about 500 mL distilled water and dilute to a volume of 1L.

B. Procedure

1. Using a sample splitter, obtain a soil sample of at least 20 g. Weigh 20.0 g of sample into a 125-mL extraction bottle.
2. Add 25.0 mL of distilled water and stir well with a glass rod to insure that the soil is completely wet. Add another 25.0 mL of distilled water to rinse glass rod and side of jar.
3. Place a sheet of parafilm, then a layer of plastic wrap over the mouth of the bottle and tightly secure the lid. Place in an incubator set at 40 plus or minus 0.5 C for 7 days (168 hr).
4. Remove samples from incubator and carefully add 50.0 mL of 2 N KCl. Replace the plastic covers and tighten lid securely.
5. Shake briskly to disperse the soil and place on a mechanical shaker for 1 hour. Filter through a Whatman No. 42 or equivalent filter paper into acid-rinsed filter vials.
6. Determine the NH₄-N content of the extract solution from the incubated sample on an automated colorimetric analyzer. This determination can also be made using the Kjeldahl distillation-titration method, described in Appendix.
7. Determine the initial NH₄-N (reference) content in the soil by following steps 1-2 and 4-6 above.

C. Calculations

$$\text{ppm mineralizable NH}_4\text{-N} = (\text{ppm NH}_4\text{-N in incubated extract} - \text{ppm NH}_4\text{-N in reference extract}) \times 5$$

D. Comments

This procedure is a modification of the anaerobic incubation described by Keeney (1982). Sample size has been increased from 5 to 20 g. A 125-mL screw-top extracting bottle is used here to accommodate the larger sample size and volume of solutions.

Because of the biological nature of this procedure, there is a higher level of variability in the results than in many other soil testing procedures. Therefore, all attempts to reduce variation are critical. To reduce experimental error, the following are recommended: thorough sample mixing, complete sealing of bottles during incubation, avoidance of floating

particles during incubation, and strict temperature control. Preliminary results showed no advantage in excluding oxygen from the headspace by introducing a N₂ atmosphere immediately prior to sealing of the incubation vessel. Keeney and Bremner (1966) reported the erratic results whenever the smell of H₂S was detected during analysis.

The mineralizable NH₄-N content of some soils has been found to vary with time in dry storage. The OSUSTL currently recommends holding samples in dry storage for a minimum of three weeks before analysis. It is also recommended that samples be rapidly air-dried at ambient temperature immediately after sampling.

WATER ANALYSIS METHODS

Irrigation Water Quality

CALCIUM, MAGNESIUM, AND SODIUM

A. Reagents

Same as used for Extractable Bases.

B. Procedure

1. Filter through Whatman No. 42 or equivalent filter paper.
2. Dilute and analyze sample filtrate following steps 3-5 of the Extractable Bases procedure.

C. Calculations

$$\text{meq of cation/liter} = \frac{\text{ppm (mg/L) of cation in sample}}{\text{meq weight of cation}}$$

BORON

A. Reagents

Same as used for soil boron test.

B. Procedure

1. Add 2 drops of CaCl₂ extracting solution to about 30 mL of the water sample. Allow to stand for 5-10 min.
2. Filter through Whatman No. 42 or equivalent filter paper.
3. Follow steps 4-9 of the Hot-Water Soluble Boron procedure for soils, substituting the water sample for the soil extract.

C. Calculations

$$\text{ppm B in water sample} = \text{ppm B in water} - \text{ppm B in yellow colored sample (if any)}$$

SALINITY

A. Reagent

1. Potassium chloride solution 0.01 N. See Soluble Salts for soils.

B. Procedure

1. Calibrate the solu-bridge with .01 N KCl by placing instrument indicator on 1.41 and turning the temperature indicator until red and green lights are of equal intensity (same as step B.4, in Soluable Salts).
2. Record the electrical conductivity reading for each sample.

pH

A. Reagents

Same as used for soil pH test.

B. Procedure

Same as used for soil pH test except use 40 mL of water sample and omit steps 1-3.

CARBONATES AND BICARBONATES

A. Reagent

1. Hydrochloric acid, 0.1 N standardized HCl - Dilute 8.3 mL of concentrated HCl to a volume of 1 L using distilled water.
2. Phenolphthalein indicator: Dissolve 0.05 g of phenolphthalein in 50 mL of 95% ethanol and dilute to a volume of 100 mL using distilled water. Mix well.
3. Mixed indicator: Dissolve 0.1 g bromocresol green and 0.02 g of methyl red indicators in 100 mL of 95% ethanol.

B. Procedure

1. Pipette 50 mL of water sample into a 125 mL Erlenmeyer flask.
2. Add 6 drops of phenolphthalein indicator.
3. Titrate with 0.1 N standardized HCl until the indicator changes from a pink color to a clear end point. If solution remains clear after addition of phenolphthalein then proceed directly to the second titration (step 4).
4. Add 6 drops mixed indicator and titrate with 0.1 N standardized HCl to a pale pink end point.

C. Calculations

1. First titration (step 3)
 $\text{meq carbonate/liter} = \text{mL of HCl} \times 2 \times \text{N of HCl} \times 20$
2. Second titration (step 4)
 $\text{meq carbonate} + \text{bicarbonate/liter} = \text{mL of HCl} \times \text{N of HCl} \times 20$

SULFATE SULFUR

A. Reagents

Reagents will be the same as for the soil SO₄-S test except that calcium phosphate solution is not required.

B. Procedure

Follow steps of the soil SO₄-S test.

C. Calculations

Determine the amount of SO₄-S from a standard curve prepared from a series of standard solutions.

TOTAL NITROGEN Kjeldahl Procedure

A. Reagents

Same used for soil TN.

B. Procedure

1. Pipette a 10.0 mL aliquot of the water sample into a 75 mL volumetric digestion flask.
2. Follow steps 2-8 of the soil Total Nitrogen procedure. The samples will be a clear blue-green color when digested. A blank should be run using 10 mL of distilled water.

C. Calculation

$$\text{ppm total nitrogen} = \text{ppm NH}_4\text{-N in filtrate} \times \frac{75}{\text{sample size (mL)}}$$

AMMONIUM AND NITRATE NITROGEN KCl Extraction Method

A. Reagents

None.

B. Procedures

1. Follow steps 2-3 of the Extractable Ammonium and Nitrate Nitrogen procedure substituting an aliquot of water sample for the KCl extract solution. The Kjeldahl distillation method requires a 50-mL aliquot of water.
2. If determinations are to be made by Kjeldahl distillation, follow the procedural steps outlined for ammonium and nitrate nitrogen in steps 3a-i.

C. Calculation

For samples analyzed with an automatic analyzer, ppm ammonium-N or nitrate-N in solution is determined directly.

NOTES

1. Distributed by Custom Laboratory Equipment, Inc., Orange City, FL.
2. The Bausch and Lomb "Spectronic 88 spectrophotometer is used in OSUSTL.
3. Some changes in the concentrations of the standard work solutions may be required to insure operation within the linear range of the spectrophotometer being used.
4. A Perkin-Elmer model 372 atomic absorption spectrophotometer is used in the OSUSTL.
5. The five-unit vacuum filtering rack used in the OSUSTL is supplied by Soil Test, Inc., Evanston, IL.
6. RD-26 Solu-Bridge, Industrial Instruments, Cedar Grove, NJ, is used in the OSUSTL.
7. A Technicon 40-position digestion unit is used in the OSUSTL (Technicon, Inc.).
8. From an unpublished procedure entitled, "A Gypsum Requirement Test, Determination of Sodium in Equilibrium Ammonium Acetate Solution," supplied by Dr. A. R. Halvorson, Extension Soils Specialist, Washington State University, Pullman.
9. In this laboratory, heating mantels and rheostat set at 90.
10. From an unpublished procedure entitled, "Procedure for Purifying Activated Charcoal," which was supplied by Dr. A. R. Halvorson, Extension Soils Specialist, Washington State University, Pullman, WA.
11. Distributed by Custom Laboratory Equipment, Inc., Orange City, FL.
12. Some changes in the concentrations of the standard work solutions may be required to insure operation within the linear range of the spectrophotometer.
13. The five-unit vacuum filtering rack used in the OSUSTL is supplied by Soil Test, Inc., Evanston, IL.
14. RD-26 Solu Bridge, Industrial Instruments, Cedar Grove, NJ, is used in the OSUSTL.
15. From an unpublished procedure entitled, "A Gypsum Requirement Test, Determination of Sodium in Equilibrium Ammonium Acetate Solution," supplied by Dr. A. R. Halvorson, Extension Soils Specialist, Washington State University, Pullman, WA.
16. All glassware should be acid washed and rinsed with glass-distilled water.
17. OSUSTL heating mantels and rheostats are set at 90.
18. From an unpublished procedure entitled, "Procedure for Purifying Activated Charcoal," which was supplied by Dr. A. R. Halvorson, Extension Soils Specialist, Washington State University, Pullman, WA.

APPENDIX*

ORGANIC MATTER Ignition Method

A. Reagents

None

B. Procedure

1. Tare a 50-mL beaker or crucible by igniting it in a muffle furnace set at 550 C, cooling it in a desiccator, and weighing it to plus or minus 1 mg (tare).
2. Place 10-20 g of air-dried soil into the tared container and place in a drying oven set at 100 C for 2-3 hr. Cool container in a desiccator and weigh (soil).
3. Place the container plus sample in a muffle furnace set at 550 C for 4-5 hr. Cool container in a desiccator and weigh (burn).

C. Calculation

$$\% \text{ O.M.} = \frac{\text{soil} - \text{burn}}{\text{soil} - \text{tare}} \times 100$$

D. Comments

This method appears to be superior to the Walkley-Black method for samples high in organic matter. However, hydrated aluminosilicates, loose structural water, and carbonate minerals are decomposed upon heating which may result in weight losses in excess of the actual organic matter content. The method outlined by Nelson and Sommers (1982) in Section 29-4.3 suggests pretreatment of the soil with a mixture of HCl and HF to remove the hydrated mineral matter. Samples containing carbonate minerals should be pretreated with HCl to dissolve all of the carbonates. To test for the presence of carbonates follow the procedure below:

Place small amount of finely ground soil on a sheet of wax paper and moisten with a few drops of water. Add approximately 4 N HCl drop-wise to the moist sample, and note any evidence of effervescence. Allow sufficient time to react.

KJELDAHL DISTILLATION CEC, TN, NH₄-N, NO₃-N and Mineralizable-N

A. Reagents

1. Mixed Indicator - Dissolve 0.3 g of bromocresol green and 0.165 g of methyl red indicators in 400 mL of 95% ethanol, and bring to 500 mL volume.
2. Boric acid indicator, 4% H₃BO₃ - Dissolve 20 g of reagent grade H₃BO₃ in about 900 mL distilled water; heat and swirl until dissolved. Add 20 mL of mixed indicator (reagent 1). Adjust to reddish-purple color or until 1 mL water added to 1 mL solution turns indicator a light green. Adjust indicator solution with 0.1 N sodium hydroxide (NaOH) (pH around 5.0) and dilute to 1 L.
3. Sodium hydroxide, 40% NaOH - Dissolve 400 g of NaOH pellets in about 500 mL distilled water. Cool and bring to 1 L volume.
4. Sodium chloride (NaCl) - Reagent grade, granular.
5. Devarda's alloy - Grind reagent grade alloy in a ball mill until it will pass a 100-mesh sieve and 75% will pass a 200-mesh sieve.
6. Magnesium oxide - Oven dry heavy magnesium oxide (MgO) in a muffle furnace at 650 C for 2 hr. Cool and store in a desiccator.
7. Hydrochloric acid, 0.1 N, standardized - Add 8.3 mL of concentrated HCl to 500 mL distilled water, then bring to 1 L volume. Standardize following the general procedure outlined in Appendix. This is used for titrations in the determination of cation exchange capacity and total nitrogen.
8. Hydrochloric acid, 0.01 N, standardized - Dilute 100 mL of 0.1 N HCl with distilled water to a volume of 1 L. Standardize following the procedure outlined in Appendix. This is used for titrations in the determination of ammonium and nitrate nitrogen.

B. Procedure

1. Turn on heating unit to boiling flask and condensers.
2. Pipette 10 mL of boric acid indicator solution into a 125 mL Erlenmeyer flask. Place the Erlenmeyer flask under the condenser tip of the Kjeldahl unit. The end of the condenser should be in the boric acid indicator. Make sure the system is boiling before attaching the Kjeldahl flask to the distillation system in Step 3.

(Note: Steps 1 and 2 precede all succeeding steps.)

* The appendix contains a combination of alternate procedures, seldom used procedures and instructions for standardization of an acid.

CEC

3. Transfer a 50 mL aliquot of leachate from CEC step 5 into a 300-mL Kjeldahl flask. Add 3 g of NaCl to leachate in flask. Place flask on system.
4. Add 20 mL of 40% NaOH to the leachate through the stopcock; rinse with a small amount of distilled water, and close the stopcock.

Note: It is advisable to turn the steam off before adding reagents through the stopcock to avoid spitting. Be sure to turn the system back on before plugging the stopcock.

5. Distill approximately 75 mL into the 125-mL Erlenmeyer flask containing the boric acid indicator. Remove the steam bypass plug and then remove the Erlenmeyer flask.
6. Titrate with 0.100 N HCl to a pink endpoint.
7. Make a blank determination following the same procedure as the samples using 50 mL of 0.1 N HCl in place of the leachate.

TN

3. Quantitatively transfer the contents of the 75-mL volumetric digestion tube into a 300-mL Kjeldahl flask and attach to distillation system.
4. Add 30 mL of 40% NaOH to the digested solution through the stop cock, rinse with a small amount of distilled water and close the stop cock. (*See Note in CEC.*)
5. Follow Step 5 in CEC distillation.
6. Titrate with 0.1 N HCl to a pink endpoint.
7. Make a blank determination on sample that was digested with each set of samples following the same procedure only without adding soil.

Extractable $\text{NH}_4\text{-N}$ and $\text{NO}_3\text{-N}$

3. Transfer a 50-mL aliquot of the filtered KCl extract solution into a 300-mL Kjeldahl flask.

$\text{NH}_4\text{-N}$ Determination

4. Add 0.8 g MgO directly to the Kjeldahl flask and immediately attach to the distillation unit.
5. Follow Step 5 in CEC distillation.
6. Titrate with 0.01 N HCl to a pink endpoint.
7. Make a blank determination following the same procedure, using 50 mL of 1 N KCl in place of the sample filtrate.

$\text{NO}_3\text{-N}$ Determination (Nitrite is also analyzed)

4. After removal of $\text{NH}_4\text{-N}$ from the sample as described in the previous section, replace the Erlenmeyer flask with one containing fresh boric acid indicator (Step 2). Then add 0.8 g of Devarda alloy through the stopcock; rinse with a small amount of distilled water and close the stopcock.
5. Follow Step 5 in CEC distillation.
6. Make a blank determination following the same procedure, using 50 mL of 1 N KCl in place of the sample filtrate.

$\text{NO}_3\text{-N}$ and $\text{NH}_4\text{-N}$ Determination

4. Follow the same procedure described for determination of $\text{NH}_4\text{-N}$, but add 0.8 g of Devarda alloy to the distillation chamber immediately after addition of MgO.

Washing of Kjeldahl distillation unit.

- a. Fill a Kjeldahl flask with 1 N HCl. Attach to the Kjeldahl distillation unit, insert the steam bypass stopcock, and turn on the steam generator unit.
- b. Allow the acid to boil over through the condenser until thoroughly flushed. Remove the plug, then remove the Kjeldahl flask.
- c. Repeat steps a and b above using distilled water.

Note: Washing is necessary to remove any traces of Devarda's alloy which may accumulate. The presence of the alloy will cause a negative error in the $\text{NO}_3\text{-N}$ determination.

D. Calculations

1. Cation Exchange Capacity in meq/100 g soil =

$$\frac{(\text{mL HCl sample} - \text{mL HCl blank}) \times \text{N of HCl} \times 5 \times 100}{\text{soil sample size (g)}}$$

2. % Total Nitrogen in soil =

$$\frac{(\text{mL HCl sample} - \text{mL blank}) \times \text{N of HCl} \times 0.014 \text{ g N/meq}}{\text{soil sample size (g)}}$$

3. ppm $\text{NH}_4\text{-N}$ or $\text{NO}_3\text{-N}$ in soil =

$$(\text{mL HCl sample} - \text{mL blank}) \times \text{N of HCl} \times 0.014 \text{ g N/meq} \\ \text{soil sample size (g)} \times \left(\frac{\text{mL of aliquot}}{\text{mL of extract}} \right)$$

E. Comments

Some of the reagents used in the Kjeldahl distillation determinations have been modified from the method presented by Bremner and Mulvaney (1982). These modifications have been developed so that the procedure can be used for routine soil analysis.

SULFATE SULFUR ($\text{SO}_4\text{-S}$) Distillation Method

A. Reagents¹⁰

1. Reducing agent - Under a fume hood, mix 400 mL of hydriodic acid (56%), 100 mL hypophosphorus acid (50%), and 200 mL formic acid (88%) in a sturdy 1000 mL beaker. Boil gently with a stream of nitrogen flowing through this solution for about 10 min after the temperature has reached 115 C. The nitrogen gas should be bubbled through the solution by passing N_2 through a glass tube placed near the bottom of the beaker. Do not let the temperature of the solution exceed 117 C. Do not attempt to recover spent reagent by distillation. Remove beaker from hot plate and maintain N_2 flow through the solution until cool. Store in glass container. Reagent is stable for two months.

CAUTION! EXTREMELY POISONOUS FUMES OF PHOSPHINE (PH₃) may be liberated from the reagent if heated above 120 C or if the reagent is spilled on a hot surface.

2. Pyrogallol - sodium phosphate wash solution (Not used unless solution contains high levels of NO₃)

a. Stock reagents

(i) Dissolve 100 g of sodium phosphate monobasic (NaH₂PO₄·H₂O) in 500 mL glass-distilled water and dilute to 1 L volume.

(ii) Crush about 100 g of crystalline pyrogallol [pyrogallolic acid, C₆H₃(OH)₃] using a mortar and pestle. Store in a tightly closed container.

b. Working wash solution

(i) Weigh 1+ g of crushed pyrogallol into a 150 mL beaker for each distillation unit to be used (e.g., 6 g for a 5-unit system).

(ii) Saturate the atmosphere in the beaker with N₂ gas. This can be accomplished by holding the end of a tygon tube from which an audible stream of N₂ gas is flowing near the bottom of the beaker for about 1 minute.

(iii) Add 12 mL of sodium phosphate monobasic solution per distillation unit to the beaker and stir with a magnetic stirrer until the pyrogallol is dissolved. An atmosphere of N₂ gas needs to be maintained above the solution to prevent the pyrogallol from being oxidized and turning yellow.

3. Zinc acetate-sodium acetate (sulfide absorbing solution) - Dissolve 50 g of zinc acetate dihydrate (Zn(CH₃COO)₂·2H₂O) and 12.5 g of sodium acetate trihydrate (CH₃COONa·3H₂O) in 500 mL glass-distilled water then dilute to 1 L volume. A bulk supply of a dilute zinc acetate-sodium acetate can be made by diluting the above solution to a 7 L volume with glass-distilled water.

4. Amino dimethylaniline solution - Dissolve 2.0 g of p-amino dimethylaniline sulfate in 1500 mL of glass-distilled water. Slowly add 400 mL of concentrated reagent grade sulfuric acid (H₂SO₄) inside cold, running water bath to cool and avoid evaporation. Dilute the cooled solution to 2 L with glass-distilled water.

5. Ferric ammonium sulfate solution - Add 15 mL of concentrated H₂SO₄ to 75 g of ferric ammonium sulfate crystals [FeNH₄(SO₄)₂·12H₂O]. Add 585 mL of glass-distilled water slowly without mixing to keep H₂SO₄ on bottom and to allow dissolution of ferric ammonium sulfate. The crystals dissolve in around 10 days.

6. Standard sulfate-S solutions

a. Standard stock solution, 100 ppm SO₄-S - Dissolve 0.5434 g of oven dry potassium sulfate (K₂SO₄) in 500 mL glass-distilled water and dilute to a volume of 1 L.

b. Standard working solutions - Prepare work solutions by pipetting the following aliquots of 100 ppm SO₄-S stock solution into 100 mL volumetric flasks (bring to volume with the appropriate potassium chloride extracting solution):

mL 100 ppm stock solution	ppm SO ₄ -S in work solution
1	1
3	3
7	7
10	10
15	15

7. Potassium chloride extracting solutions -

a. Eastern Oregon: 1 N KCl - Dissolve 74.56 g potassium chloride (KCl) in 500 mL of glass-distilled water and dilute to 1 L volume.

b. Western Oregon: 1 N KCl + KH₂PO₄ - Dissolve 4.39 g KH₂PO₄ and 74.56 g KCl and bring up to 2 L with glass-distilled water.

8. Nitrogen gas (prepure)

9. Sulfur-free ground joint lubricant - Most ground joint lubricants contain appreciable sulfur that must be removed before use. Many lubricants deteriorate quickly when exposed to the hot reducing agent. Dow-Corning silicone stopcock lubricant has been found suitable if freed from sulfur contaminant. Place about 5 g of the silicone lubricant in a 100-mL beaker, add 5 mL of hydriodic acid and 5 mL of hypophosphorous acid. Place a watch glass filled with distilled water on top of the beaker to act as a condenser. Boil the mixture gently with frequent stirrings for about 45 min. Allow to cool, pour off the acid mixture, and wash the lubricant thoroughly with glass-distilled water.

B. Procedure

1. Extraction of SO₄-S

a. Weigh 10 g of soil into a 50 mL plastic bottle.

b. Add 20 mL of the appropriate KCl extracting solution and shake for one hour. The shaking action should be sufficiently vigorous to keep the soil suspended in solution.

c. Filter through Whatman No. 42 filter paper (or equivalent).

2. Preparation of digestion-distillation apparatus

a. Rinse washing columns with 0.5 N NaOH and then glass-distilled water.

b. Lubricate all spherical joints with a minimal amount of S-free lubricant.

c. Saturate the column with N₂ gas to reduce the possibility of oxidizing the pyrogallol. Place 10 mL of the pyrogallol-sodium phosphate wash solution in the gas washing column, then resaturate the column and solution with N₂ gas. Plain water may be used in gas traps unless solutions contain high levels of nitrate. Reattach the columns to the apparatus.

d. Saturate the system (digestion-distillation apparatus and washing solution) with H₂S by using a 15 ppm SO₄-S standard solution. Follow sulphur determinate described below with the following exception: Vent H₂S-N₂ into the hood when the system is being saturated.

Note: Saturation should be done prior to analyzing samples each day or when new solution is introduced. The solution should be changed when yellow color appears or when the system has been used 25-30 times.

3. Determination of $\text{SO}_4\text{-S}$

a. Place 50 mL of the dilute zinc acetate-sodium acetate solution into a 100-mL volumetric receiving flask. Connect the glass delivery tube to the side arm of the gas washing column. Place the receiving flask with the delivery tube inside and near the bottom of the receiving flask, but not touching it.

b. Pipette a 2.0 mL aliquot of standard solution or sample extract into a 50 mL digestion-distillation flask and add 4 mL of reducing reagent. It is recommended that this and all succeeding steps (3b through 3h) be conducted under a suitable fume hood.

c. After moistening joint with a drop of water to insure a complete seal, immediately attach the digestion-distillation flask to the condenser and connect the nitrogen supply tube. Adjust the N_2 flow rate to about 2 bubbles per second. Make certain cool water is passing through the condenser.

d. After 5 min of N_2 flow to obtain a reduced atmosphere, apply heat to the digestion-distillation flasks by either lighting suitable microburners or positioning preheated heating mantels around the base of the flask. With N_2 still flowing, heat the contents of the flask and maintain at a low boil¹¹ for one hour.

e. Remove the receiving flask, leaving the glass delivery tube in the zinc acetate solution. Immediately add 10 mL of the amino dimethylaniline solution. Quickly stopper the receiving flask and mix thoroughly.

f. Add 2 mL of ferric ammonium sulfate solution and shake. Allow blue color to develop for at least 1/2 hr but no longer than 10 hr. Dilute to a 100 mL volume with glass-distilled water and mix thoroughly, leaving glass tube inside.

g. The blue color developed will be quite stable after 30 min. and should be read within 24 hr on a suitable spectrophotometer set at 670 nm.

h. Prepare standards following steps 3a-g, substituting 2.0 mL of the standard work solutions for the soil extract. A blank is prepared in the same manner using 2.0 mL of the appropriate extracting solution instead of soil extract.

i. If the color is more intense than that obtained for the highest standard work solution, make an appropriate dilution. For best results, dilute the soil extract to a concentration within the linear range of standard work solutions using the appropriate KCl extracting solution and following steps 3a-g.

C. Calculations

$$\text{ppm } \text{SO}_4\text{-S in soil sample} = \text{ppm } \text{SO}_4\text{-S in soil extract} \times 2$$

D. Comments

The methylene blue method for the determination of sulfur as described by Tabatabai (1982) is followed except for the following modifications:

1. A special technique is used to make up the pyrogallol-sodium phosphate wash solution. When the wash solution is prepared in the manner described above, up to 25 determinations can be made before the solution becomes discolored.

2. The zinc acetate-sodium acetate is made up in the dilute form.

The methylene blue method used here yields more accurate values than the turbidimetric procedure of Tabatabai and Bremner (1970). A modified turbidimetric method has also been used for sulfur analysis but is not described here.

CALCIUM CARBONATE EQUIVALENT FOR LIMING MATERIALS AND HIGHLY BASIC SOILS

A. Reagents

1. Hydrochloric acid, 0.500 N HCl, standardized - Dilute 46.5 mL concentrated HCl to a volume of 1 L with distilled water. Standardize against 25 mL of 0.500 N sodium carbonate (Na_2CO_3) or sodium hydroxide (NaOH). These standard base solutions are available through most chemical suppliers, or can be prepared from pure, dry reagent Na_2CO_3 or NaOH.

2. Sodium hydroxide, 0.500 N NaOH, standardized - Dissolve 20.00 g NaOH pellets in about 500 mL distilled water. Cool and dilute to a volume of 1 L. Standardize against the 0.500 N standard HCl (reagent 1).

3. Phenolphthalein indicator - Dissolve 0.05 g phenolphthalein in 50 mL of 95% ethanol. Bring to 100 mL volume with distilled water.

B. Procedure

1. Place 1.0 g of ground liming material or 5 to 10 g of soil in a 150-mL Erlenmeyer flask. To initially determine how much soil to use, add a drop of 0.5 N HCl to some of the soil. If the soil effervesces, 5 g should be used.

2. Add 50.0 mL of the standardized 0.5 N HCl to the Erlenmeyer flask and boil gently for 5 min. A watch glass filled with cool distilled water placed on top of the flask will act as a condenser.

3. Allow the solution to cool. Rinse any condensation on the watch glass into the solution with distilled water. For soil, filter through a Whatman No. 42 or equivalent filter paper into a 250-mL flask, washing all soil from the Erlenmeyer flask with distilled water.

4. Titrate the excess acid with the standardized 0.5 N NaOH, using 4 drops of phenolphthalein indicator. The end point will be pink.

C. Calculations

$$\% \text{ calcium carbonate equivalent} = \frac{(\text{mL of HCL} \times \text{N of HCL}) - (\text{mL of NaOH} \times \text{N of Na OH}) \times 0.05}{\text{sample size (g)}}$$

D. Comments

The above test should be used for materials with percent calcium carbonate greater than 20. If percent calcium carbonate is less than 20, use the carbonate method found on p. 12. The above method does not differentiate between calcium and magnesium carbonates.

STANDARDIZATION OF ACID

A. Reagents

1. Sodium carbonate, 0.1 N (Na_2CO_3)
2. Acid - Acid of unknown normality to be standardized.
3. Mixed indicator - Dissolve 0.1 g of bromocresol green and 0.02 g of methyl red indicators in 75 mL of 95% ethyl alcohol, then bring to 100 mL volume.

B. Procedure

1. Pipette a known amount of 0.1 N Na_2CO_3 into a 100-mL beaker.

Note: Use 10 mL for acid around 1.0 N, and 1.0 mL for acid around 0.1N.

2. Add 5 drops of mixed indicator.
3. Titrate with the unknown acid to a pink endpoint.
4. Calculate the normality of the acid.

C. Calculation

$$\text{Normality of acid} = \frac{(\text{N of Na}_2\text{CO}_3) (\text{mL Na}_2\text{CO}_3)}{\text{mL of acid used to titrate}}$$

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Appendix 3B
Quality Control At OSU Soil Testing Laboratory



OREGON STATE UNIVERSITY

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Tel: 503-737-2441 · Fax: 503-737-3178

Quality Control at OSU
Soil Testing Laboratory
February 20, 1991
By Donald A. Horneck

It is the policy of the OSU Soil Testing Laboratory (OSU-STL) to take the necessary steps to insure that quality results are maintained. This is done in many ways in the laboratory. First of all the OSU-STL participates in a northwest regional sampling program operated through Utah State University. This program allows us to compare our results with other laboratories throughout the region. For data regarding results from this program please contact the Soil Testing office.

General

To insure unbiased analysis and accurate record keeping, samples are assigned a unique lab number, which is written with a permanent marker on each sample bag. Batches of samples are brought up on trays of 32. The first and 16th soil samples on each tray are internal lab standards. The 32nd sample on each tray is duplicated. Samples on each tray are given a consecutive number to minimize the potential of samples getting out of order. Glassware is arranged and numbered in racks of eleven, three racks to a tray.

Soil reference samples are collected, ground and mixed. They are analyzed with a previously established reference sample to determine values. The results from the reference samples are recorded every time they are analyzed and kept on file. Tolerances are set, generally one standard deviation. Reference samples are evaluated when a batch of samples is run. The reference sample is used as a way of insuring that samples are in their correct order and that procedures are operating correctly.

More information as to how instruments are calibrated, samples are prepared and solutions are mixed can be found in our methods manual.

Bases - K, Ca, Mg, Na

Several steps are taken to insure accurate results. The instrument (Perkin Elmer 372) is calibrated every time it is used and when elements are changed. The five point plus a blank standard curve is recorded so that day to day fluctuations are known. When running a batch a point on the curve is checked every 11 samples with the whole curve checked after every tray (33 samples).

Samples are diluted with a lanthanum and lithium solution which minimizes interferences and gives a uniform salt background. Lanthanum is added to eliminate interferences with calcium and magnesium. Lithium is added to prevent ionization of sodium and potassium.

Standards are mixed from purchased solutions that are traceable to NBS standards and diluted in the same way that samples are handled. Reference samples are run every 15 samples and a duplicate every 30 samples.

Phosphorous (Bray)

Phosphorous is run on a continuous flow analyzer (Alpkem RFA) using the molybdate blue method with an in line-dialyzer. A standard curve consisting of four points and a blank is run every 35 samples. A constant check is maintained on baseline drift. Multiple sampling is done where increased precision is needed. Reference samples are analyzed every 15 samples and a duplicate every 30 samples.

Constant shaking times are maintained. Colloidal contamination is visually evaluated after filtration and samples are refiltered when necessary.

Organic Matter or Carbon (OM, OC)

Samples are hand ground to pass a 0.50 mm sieve to insure that fresh organic material is excluded and help increase surface area for reaction. Normality of the titrant is checked (blank) every 20 samples. Reference soil samples are analyzed with every blank and recorded.

pH

Samples are scooped and read in exact tray order. The pH meter is calibrated with purchased buffer solutions that are traceable to NBS standards. Reference samples are run every 15 samples and a duplicate every 30 samples.

Nitrates and/or Ammonium

Samples are weighed into numbered bottles. Filter paper is leached first with 50-100 ml KCl prior to filtration of sample to minimize contamination from filter paper. Reference samples and blanks are analyzed a minimum of every 25 samples.

Nitrate and ammonium are run on a continuous flow analyzer (Alpkem RFA) using cadmium reduction and indophenol methods, respectively. An in line-dialyzer is also used. A standard curve consisting of four points and a blank points is run every 35 samples. A constant check is maintained on baseline drift. Multiple sampling is done where increased precision is needed. Reference samples are analyzed every 15 samples and a duplicate every 30 samples.

Constant shaking times are maintained. Colloidal contamination is visually evaluated after filtration and samples are refiltered when necessary.

Figure 1 table 2. Project QA Organization

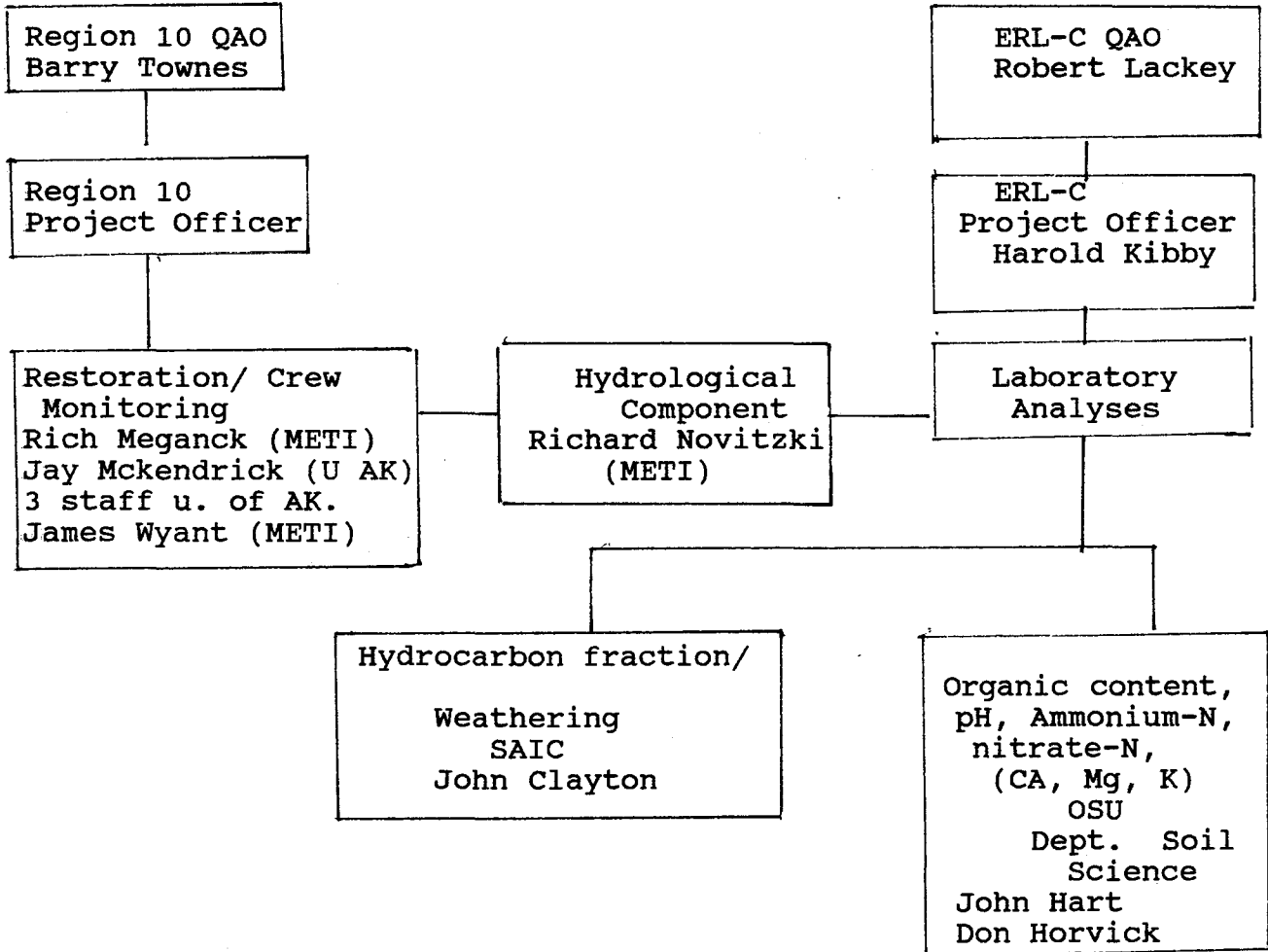


Table 4. Example of Corrective Actions for Operation of Atomic Absorption Spectrophotometer


Type of QC Check	Frequency	Precision	Accuracy	Corrective Action
Blank	At beginning of calibration or recalibration		0.00	Check concentration of purified water. Check instrument operating conditions.
Calibration standards	At beginning of each batch analysis		$r^2 \geq 0.98$	Rezero and recalibrate, verify calibration standard values. Check instrument operating conditions. Change O-ring on nebulizer.
Low concentration QCCS	2X minimum in batch	5%	98% recovery	Replace lamp. Reanalyze 2nd duplicate, rezero and recalibrate.
High concentration QCCS	2X minimum in batch	5%	98% recovery	Reanalyze 2nd duplicate, rezero and recalibrate.
Sample duplicates	1 in every 15 routine samples	7%		Reanalyze 2nd duplicate. Reanalyze last 15 samples.
NBS standard*	2X in every batch	5%	1% from certified value	Reanalyze 2nd NBS sample. Rezero and recalibrate.

QC Quality Control
QCCS Quality Control Check Sample
NBS National Bureau of Standards
* or other certified reference material

Figure 2. Agreement to Comply

My signature below indicates that:

1. I have read the QA project plan for the project "Feasibility of Restoring the Bay of Isles and Tonsina Bay in Prince William Sound and the Gulf of Alaska".
2. I have read the QA procedures that are unique to my project activities (Sections 6, 7, 8, 9, 10, 11, 13, 14, 15, 16, 17).
3. I accept the responsibility of adhering to the procedures outlined in this QA document.



Signature

2/28/91

Date

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RPWG
N

Title: Population Assessment of the Prince William Sound Sea Otter Population

Submitted to: Restoration Planning Work Group

Principal Investigators: James L. Bodkin and Mark S. Udevitz

Study Period: 1, March 1991 to 1, March 1994

Lead Agency: U.S. Fish and Wildlife Service

Cost of Proposal: First Year; 176.6K

Cost Allocation: R7 Oil Spill Restoration 150.0K
R8 Sea Otter Research Project 26.6K

Date of Plan: 27, February 1991

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Anchorage, Alaska

DRAFT

1. WORK UNIT TITLE: Interactions Between Sea Otters and Fisheries in Alaska

2. STUDY PLAN TITLE: Population Assessment of the Prince William Sound Sea Otter Population

3. ORGANIZATION: 87200 Alaska Fish and Wildlife Research Center

4. WORK UNIT: 00024

5. SECTION: 210.03

6. PRINCIPAL INVESTIGATORS: James L. Bodkin and Mark S. Udevitz

7. PROJECT COSTS: FY 1991 26.6K
(150K additional support from R7 Restoration funding; Total year 1 cost: 176.6K)

8. SIGNATURES:

Project Leader: _____ Date: _____

Branch Chief: _____ Date: _____

Director, AFWRC: _____ Date: _____

DRAFT

1. INTRODUCTION

Initial damages to the sea otter population resulting from the T/V Exxon Valdez oil spill included lethal and sub-lethal levels of direct exposure. One method used to estimate the total immediate loss to the sea otter population in Prince William Sound was a comparison of estimates of sea otter abundance based on boat surveys conducted before and after the spill. Boat surveys were used to estimate the population size in 1989 and 1990 in order to be consistent with the method used before the spill. This consistency was necessary for damage assessment, but it has become evident that the boat survey methodology as conducted will not provide the accuracy or precision necessary to monitor changes that may occur in the post-spill population.

Changes that may occur in the western Prince William Sound sea otter population following the initial mortality resulting from exposure to oil are unknown. Initial and chronic, sub-lethal exposure of sea otters to hydrocarbons through the environment or their prey base may cause population losses that will not become evident for many years. Alternatively, the removal of a large percentage of the western Prince William Sound sea otter population by the spill may release the survivors and immigrants from density dependent factors regulating sea otter abundance. It is likely that annual changes in post-spill abundance will be of a lesser magnitude than initial spill induced mortality.

Methods used in the past to obtain estimates of sea otter abundance include counts from the ground (Estes and Jameson 1988), small and large vessels (Jameson et al. 1982) and fixed (Ebert 1968, Simon-Jackson et al. 1986) or rotary wing (Douglas et al. 1990) aircraft, or a combination of two or more methods. Ground counts probably provide the most accurate estimates of nearshore sea otter abundance (Schneider 1971). Estes and Jameson (1988) estimated a sightability of 94.5% for standardized ground counts. However, ground counts have limited application throughout most of the species' range.

Sea otter surveys conducted by boat such as those conducted in Prince William Sound also have limited application within the range of sea otter habitat, although Schneider (1971) felt they provided higher counts than aerial surveys. Udevitz et al. (1990) determined that detection of sea otters in boat based surveys is reduced due to avoidance behavior of the otters as well as sightability problems. In addition to surveying sea otter abundance, boat surveys have been used to provide indices of reproductive rates in sea otter populations (Estes 1990).

Preliminary studies reported by Douglas et al. (1990) suggest that rotary-winged aircraft might be suitable as an observation platform. Schnieder (1971) suggests that rotary-winged aircraft surveys may provide counts two to four times greater than fixed-wing aircraft. However, cost-efficiency and safety considerations

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of rotary-wing aircraft over water may preclude their wide spread use in sea otter surveys.

Fixed-winged aircraft may be the most cost effective and broadly applicable survey platform for sea otters. A float equipped, single engine aircraft should provide the necessary safety factor for surveying sea otter habitat that may occur considerable distances from shore. Aerial counts of sea otters have been used for several decades throughout their range. Kenyon (1969) felt aerial surveys provided higher counts than those obtained from small boats. Traditionally, fixed-winged surveys have been conducted without standardized protocols, using different aircraft (eg. DC-3, Grumman Goose) and procedures (eg. airspeed, altitude, number of observers, weather conditions, area surveyed, etc.). Aircraft speed has been identified as one of the most important variables in defining the probability of detection of an animal from the air (Caughley 1974). The Piper PA-18 "Super-Cub" has been selected repeatedly for wildlife survey work based on its slow stall speed and high degree of maneuverability (Erickson and Siniff 1964, LeResche and Rausch 1974, Gasaway et al 1986). It seats one pilot and one passenger in tandem, an arrangement recommended by Erickson and Siniff (1964) as allowing navigation and observation to occur from the same spatial orientation in the plane.

Line transect and strip transect methods are widely used in aerial surveys to estimate population size. They rely on being able to observe all of the animals in some region (eg. on the line or in the strip) with a probability of 1.0. Due to otter behavior this is not possible using standard line or strip transect methodologies. To obtain valid population estimates, the standard methodologies would have to be supplemented with a technique to provide an estimate of the actual detectability of animals in some region covered by the survey. Our approach would be for the plane to conduct circling maneuvers along the transect at intervals, searching a specified area at a level of intensity necessary to observe all of the otters and thus obtain an estimate of the proportion seen with the standard methodology. This approach depends on the ability to actually observe all of the otters in a specified area at some level of search intensity. We will evaluate the effect of this search intensity on sea otter detectability.

Another method of aerial survey we will evaluate, in terms of application to sea otters, involves estimating population size based on systematically searching relatively large segments of habitat within a study area. This method, described by Gasaway et al. (1986) has been successfully used for estimating moose population sizes from aerial surveys. The method involves an aerial search of sample units of moose habitat at a defined search intensity. A sub-sample of these units is searched at a higher intensity at which it is assumed that all moose are observed. We will evaluate the effect of search intensity on sea otter detectability using Gasaway et al. (1986) type search patterns. If it is possible to observe all of the otters in a sample unit with some search intensity, then we would be able to use that intensity

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on a sub-sample of units to adjust for detectability bias in a full survey.

The purpose of this study is to develop a survey methodology that will provide unbiased estimates sea otter population size and is applicable throughout the species' range. This will require a quantitative evaluation of factors contributing to detectabilities of less than 1.0 for sea otters from potential survey platforms. We will initially conduct a ground-truth study to assess sea otter detectability from a fixed-wing aerial platform under different search patterns, intensities and altitudes. Replication will provide information on variability over time and space to be used in survey design. If the aircraft and methodology tested in the first year proves to be effective, then information from the first year will be used to design a survey to be implemented in the second and third years. Otherwise, additional platforms and/or methodologies will be tested in the second year with the intent to design and implement a survey in the third year. Following development of the necessary methodologies, surveys will be initiated to monitor changes in the abundance of the Prince William Sound sea otter population and describe patterns of habitat use within the Sound.

Another purpose of this study is to begin a systematic survey of the western Prince William Sound sea otter population by boat to document annual rates of reproduction. To accomplish this, we will design and implement a survey to provide annual estimates of pup to non-pup ratios for subpopulations in oiled and non-oiled portions of the western sound. Possible long term effects on reproduction will be monitored by comparing pup ratios in oiled and non-oiled regions.

Objectives:

1991

- 1) Evaluate the feasibility of using the Piper PA-18 aircraft as a sea otter survey platform.
- 2) Design and implement a small boat survey to estimate pup to non-pup ratios in oiled and non-oiled portions of western Prince William Sound.

1992

- 1) Develop a procedure for estimating the abundance of sea otters in Prince William Sound.
- 2) Implement a sea otter survey method in Prince William Sound.
- 3) Monitor reproduction in the sea otter population in western Prince William Sound.

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1993

- 1) Monitor trends in sea otter abundance and distribution in Prince William Sound.
- 2) Monitor reproduction in the sea otter population in western Prince William Sound.

This study plan addresses only 1991 objectives. Detailed study plans for 1992 and 1993 objectives will be developed, based on the results of this study plan.

Hypotheses

1. H_0 : Altitude of survey platform has no effect on sea otter detectability.
2. H_0 : Search intensity has no effect on sea otter detectability from an aerial platform.
3. H_0 : The search pattern of an aerial platform has no effect on sea otter detectability
4. H_0 : The pup to non-pup ratio does not differ between oiled and non-oiled areas in western Prince william Sound

Data Needs

The following data are critical to accomplishing objectives:

1. Detectability of sea otters as a function of survey altitude.
2. Detectability of sea otters as a function of search intensity.
3. Detectability of sea otters as a function of survey search pattern.
4. The number of sea otter pups and non-pups in a sample from oiled and non-oiled sea otter habitat in western Prince William Sound.

2. METHODS

Study Design

Year one study design will incorporate a replicated Latin square design controlling for day and site, conducted in two phases. Phase one will measure the effect of altitude and search intensity

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for the circling maneuver on a line transect. Altitudes will be 150 ft., 250 ft. and 350 ft. Each of three survey units will be flown at each of the altitudes over a three day period, with 1 flight per unit per day. The design will be replicated twice. Intensity of the circling maneuver will increase from five circles (diameter = 0.5 km) until five minutes of circling reveal no additional sea otters. Circle locations will be indicated by a marker established by the ground crew at a point on the circumference of the circle. Phase one should occur in May of 1991. Results of Phase one will provide direction in completing the design of Phase two.

Phase two will measure the effect of four search methods on sea otter detectability. These will consist of Gasaway et al. (1986) type search patterns at three intensities and the transect pattern with circling maneuvers. The altitude of these flights will be determined based on the results of phase 1. Each of 4 survey units will receive each of the 4 methods over a four day period, with one method per unit per day. The design will be replicated 4 times. Ground circles will be located by a marker established by the ground crew at a point on the circumference of the circle. Phase two should occur in July of 1991.

Study Site Selection

Survey segments will be selected in Prince William Sound based on the presence of sea otters and the lack of canopy forming kelps. These survey segments must be adjacent to an accessible vantage point with an elevation greater than 3m that offers unrestricted visibility over the site. The size of each segment will be flexible, based on the observation team's ability to count and locate with complete confidence all sea otters observed within its boundaries. Boundaries will be defined using prominent geographical features of the coastline such as offshore rocks, points of land and coves or bays. These boundaries will be accurately drawn on charts identical to those used by the air survey crew. The offshore boundary will be determined with range finders and navigational charts.

Ground Truth Crews

Ground truth crews will consist of two members each. At least one member of each team will have extensive experience in observing and counting sea otters with the use of Questar telescopes and binoculars, and will serve as the primary observer. The second team member will have some experience and training in sea otter observation. Ground observations will follow protocols established by Estes and Jameson (1988).

Ground Truth Procedures

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Each crew will be transported to a survey segment at least one hour prior to the arrival of the aerial survey crew. The ground crew will approach the ground truth segment after a thorough study of the area from offshore to locate sea otters within the survey area. The ground crew, aboard the transport vessel, will approach the coastline in a manner to minimize disturbance to sea otters. Ground crews will be deployed outside the survey area and will walk into the observation site as far from the shore as possible. Every effort will be taken to minimize disturbance to the survey area. Timing of survey crew deployment will allow each crew a minimum of one hour to locate and map the position of each otter in the survey area. Ground crews will be responsible for defining the boundaries of their unit and establishing a circumference point for the aerial circling maneuver prior to the arrival of the survey craft. At 15 minute intervals and once immediately prior to survey craft arrival, ground crews will record the location, group size, and activity of each otter or group of otters. Activity categories will include resting, grooming, foraging, swimming or hauled out. High quality range finders will be used to determine the distance to each otter and distance to the unit boundaries. Ground crews will also record the location and behavior of all otters observed outside the boundaries of the unit, observations regarding changes in sea otter activity associated with the approach of the survey craft, the time the survey craft enters and departs the segment, and environmental conditions (wind speed/direction, cloud cover and tidal level and sea state).

Following the departure of the survey crew, the ground crew will call via hand held VHF radio for the transport vessel to pick them up and transport them to the next site. It is anticipated that each ground crew can survey two or more ground truth segments per day.

Aerial survey methods

Two search patterns will be evaluated. In one, the plane will follow a linear path along the edge of the survey segment, mapping the locations of all observed otters in the segment. At intervals, the aircraft will circle on a .5 km radius inside the line. The locations of any additional otters observed within this radius will be mapped separately. Search intensities will be increased by increasing the amount of time spent circling.

The other search pattern will cover the entire segment following the general guidelines of Gasaway et al. (1986). The pattern we use will depend on the shape of the segment, but will generally consist of a rolling spiral. Search intensity will be varied by decreasing the radius of the spirals and therefore increasing the time spent in the segment. The locations of all observed otters in the segment will be mapped.

Caughley (1974) identified altitude as an important variable in defining detectability from aerial surveys. Altitude will affect detectability in both of these approaches. As altitude increases,

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above some minimum, sea otter detectability should decrease. At low altitudes, habitat will pass by too quickly to adequately observe. Also, at low altitudes, avoidance behavior may become a problem. We will evaluate the effect of altitude on detectability to indicate the optimum altitude for surveys. Typically, aerial surveys of sea otters have been conducted from about 150 ft. to 300 ft. above sea level (Schneider 1971). We will evaluate the effect of this variable on detectability at three altitudes (150 ft., 250 ft., and 350 ft.).

Several environmental variables are believed to effect the detectability of sea otters (Schneider 1971). These include time of day, wind velocity, wave height, swell height, cloud cover, precipitation, glare and elevation of the sun. While we will be unable to control for these variables we will only conduct surveys under those conditions considered to create good to excellent observational conditions, eg. wind < 8 knots, little or no surface chop, or poor visibility as outlined by Schnieder (1971) and Estes and Jameson (1988).

Aircraft speed during the surveys will be maintained as close as possible to 55 mph. Otter detectability may be greatest during the mid-day period when they are resting rather than actively feeding. Survey activities will begin when ground observations indicate that most otters have resumed normal behavior following deployment of the ground crews and will continue throughout the day. Information on variability in detection in time and space will be necessary for the efficient design of a population survey. Flights throughout the day will be used to assess the effect of the diurnal activity pattern of sea otters on detectability.

Reconciliation

At the end of each survey day, ground and aerial crews will compare the mapped locations of all observed otters. For the otters present in segment i , $i=1, \dots, r$, when the craft arrived, the number observed by both crews (b_i), the number observed only by the ground crew (g_i), and the number observed only by the survey craft (s_i) will be determined. The number of otters in the segment before any response to the approaching survey craft (a_i) will be determined based on ground crew observations prior to the arrival of the survey craft. Resolution of apparent differences between aerial and ground maps will be facilitated by the recorded times and activities.

Reproduction

Estimates of annual reproduction, as indicated by pup to non-pup ratios of sea otters will be obtained from small (<10m) boat surveys. Surveys will be conducted once per year, in June or July, following the peak pupping period. Surveys will incorporate a random sampling design within the heavily oiled and non-oiled areas of western Prince William Sound. Each area will be divided into

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sample units of sea otter habitat from which samples will be drawn.

Sampling methods will consist of classifying each sea otter observed within each sample unit as either a dependent pup or a non-pup. Dependent pups will be defined as a sea otter smaller than an adult and in close association with an adult. This definition will include, but not be limited to, a pup in close physical contact, nursing, receiving food from, swimming with or being groomed by an adult sea otter. Non-pups will be defined as all other sea otters.

Surveys crews will consist of two observers, including the boat operator. Boat speed will be less than 15 mph. Surveys will be conducted only when viewing conditions are considered good or better (calm to light winds, sea state less than Beaufort 2). Observers will each have the use of high quality binoculars. Each otter or group of otters will be approached as close as necessary to accurately classify each sea otter as either pup or non-pup.

Boat surveys will be conducted once per year following the peak of pupping to develop a ratio of the number of pups per non-pups in the population. This ratio will be developed by observing a sample of about 400 sea otters from close range and determining the presence or absence of a pup or pups, in each of the study areas. This survey will be conducted once per year and will be conducted at the same time in subsequent years.

Safety requirements:

All staff scheduled for aircraft survey work will have current basic aircraft safety, first aid, CPR and survival courses.

All staff scheduled for field work will have current first aid, CPR and survival courses.

3. INFORMATION REQUIRED FROM OTHER INVESTIGATORS

The successful completion of this study may depend on available information from past boat surveys of sea otters in Prince William Sound describing distribution and abundance.

4. DATA ANALYSIS

Estimation of detectability and its component probabilities will follow Udevitz et al. (1990). The proportion of the otters that leave the segments in response to the approaching aircraft and are therefore not available to be counted (avoidance probability) will be estimated as

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$$\hat{P}_a = \frac{\sum_{i=1}^r (a_i - b_i - s_i - g_i)}{\sum_{i=1}^r a_i}$$

with variance estimated as (following Cochran 1977; pg. 305)

$$\text{Var}(\hat{P}_a) = \frac{\sum_{i=1}^r (-b_i - s_i - g_i + a_i(1 - \hat{P}_a))^2}{r(r-1)\bar{a}^2}$$

where

$$\bar{a} = \frac{\sum_{i=1}^r a_i}{r}$$

The proportion of the otters remaining in the segments while they are surveyed that are observed by the air crews (sighting probability) will be estimated as

$$\hat{P}_s = \frac{\sum_{i=1}^r b_i}{\sum_{i=1}^r m_i}$$

with variance estimated as

$$\text{Var}(\hat{P}_s) = \frac{\sum_{i=1}^r (b_i - m_i \hat{P}_s)^2}{r(r-1)\bar{m}^2}$$

where $m_i = b_i + g_i$ and

$$\bar{m} = \frac{\sum_{i=1}^r m_i}{r}$$

The proportion of the otters present in the segments before any response to the aircraft that are observed by the air crews during the survey (detection probability) will be estimated as

$$\hat{P}_d = \frac{\sum_{i=1}^r (b_i + s_i)}{\sum_{i=1}^r a_i}$$

with variance estimated by

$$\text{Var}(\hat{P}_d) = \frac{\sum_{i=1}^r (b_i + s_i - a_i \hat{P}_d)^2}{r(r-1)\bar{a}^2}$$

The sighting probability estimate is equivalent to the Peterson-type estimates used by Magnusson et al. (1978) for aerial surveys of crocodile nests and modified by Estes and Jameson (1988) for

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ground based surveys of sea otters. This estimate is based on the following assumptions:

1. The presence and activities of the ground crews do not affect the distribution or sightability of the otters.
2. The probability of observing an otter from shore is independent of the probability of observing it from the air.
3. Ground crews are able to make accurate determinations of ground truth segment boundaries and the locations of otters within those segments immediately before they exhibit any response to the aircraft.
4. Comparisons of otter location maps produced by ground and air crews will provide accurate determinations of b_i , g_i , and s_i , $i=1, \dots, r$.

The estimates of avoidance and detection probabilities are extensions of these that assume all otters observed in a segment by an air crew were among the otters observed by the ground crew on that segment before or during the time when the aircraft was present. The detection probability represents the overall proportion of the otters detected by the air survey. It accounts for otters that leave the segment in response to the aircraft before the aircraft arrives as well as otters that are in the segment when the aircraft arrives but are not observed.

Latin square analysis of variance with appropriate transformation of the dependent variable will be used to assess the effects of altitude, pattern and intensity on detectability. The test for altitude effect will be based on the maximum detectability obtained during each set of circling maneuvers. The optimum search intensity for the transect-circling pattern and the optimum altitude for both search patterns will be determined from phase 1 data. The circling maneuver is assumed to be similar enough to the maneuvers used in the Gasaway et al. (1986) search patterns, that the effect of altitude will be the same for both methods. Methods will be evaluated based on the altitude and intensity combination resulting in the highest estimated detectability. Phase 2 will compare the transect-circling pattern selected on this basis with Gasaway et al. (1986) patterns at three intensities. Application of any of these approaches to sea otter surveys will rely on the assumption that all otters in specified circles or units are detected, using the optimum altitude and intensity combination. We don't expect this assumption to be strictly valid on every occasion, but will accept that it will be reasonable for practical purposes if the estimated detection probability is .95 or greater.

5. SCHEDULE

March through April 1991: Evaluate available literature on wildlife census procedures, particularly that pertaining to sea otters.

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May through October 1991: Conduct initial field tests of the PA-18 survey platform. Conduct reproduction survey.

November through February 1992: Analyze and prepare findings.

1992 Implement census procedure, dependent on satisfactory results in FY1991, refine methodology, test procedures at second location. Continue reproduction surveys.

1993 Continue implementation and refinement of procedures. Continue reproduction surveys.

6. ANIMAL HEALTH AND WELFARE

We do not anticipate the handling of live animals in this project. Disturbance to animals in the wild will be minimized. Activities will be discontinued if large scale influence on animal behavior is observed.

7. STAFFING

Staffing requirements will be met by the principal investigator, project staff, FWS cooperators, OAS and private contractors as necessary.

8. LOGISTICS

Study implementation will depend on close coordination between ground and aerial crews. Field camps will be established to supply basic services for staff. Fuel will be purchased in bulk and transported to fuel caches prior to the initiation of field work. Travel to and between study sites will be by suitable vessels (eg., 25' Boston Whalers and Super Cub aircraft).

9. BUDGET

A. Costs

Line

100	200	300	400	500	Total
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Year 1 Projected Expenses 78.7K 18.1K 64.2K 8.6K 7K 176.6

Year 1 allocations: 150K Restoration
26.6K 87200-1411

Projected Expenditure Breakdown

Line 100 - Salaries (does not include full-time permanent

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staff, 6 months of staff time are being paid under other studies.

<u>Grade</u>	<u>Name</u>	<u>Monthly Salary and Benefits</u>	<u>FTE</u>	<u>Person Months</u>	<u>Costs</u>
GS 9	staff	\$3,192	1	12	\$38,304
GS 7	staff	\$2,300	1	12	\$27,600
GS 9	staff (non-staff field crew time)	\$3,192	0	4	\$12,768
Total					\$78,672

<u>Position</u>	<u>Location</u>	<u>Time Frame</u>	<u>Person Months</u>
GS 9	Office	3/91-4/91	2
GS 9	Field	5/91-10/91	15
GS 9	Office	11/91-2-92	4
(GS 9 field includes 4 non-staff months and 5 non-study staff months)			
GS 7	Office	3/91-4/91	2
GS 7	Field	5/91-10-91	6
GS 7	Office	11/91-2/92	4

Line 200 - Travel

	<u>Costs</u>
In state Travel and per diem	
Travel in state (Whittier, train costs)	\$2,300
10 trips to Cordova @ 400 ea. (inc. charter)	\$4,000
per diem, AK 30 days @ 125/day	\$3,750
Travel CA to AK, 4 @ 2K	\$8,000
Total	\$18,050

Line 300 - Commodities

A. Food and supplies, (\$20/day X 450 days)	\$9,000
B. Boat Fuel, 80 gal/day X 30 days X 2 boats X \$4 gal ea	\$19,200
C. Aviation Fuel 200 hrs @ 7 gal/hr X \$4/gal	\$5,600
D. Rangefinders, 4 @ \$100 ea	\$400

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E. Super Cub charter, 200 hrs @ 150 ea \$30,000

Total \$64,200

Line 400 - Equipment

A. Questar Telescopes, 1 ea \$3,000

B. Binoculars 4 @ 900 ea \$3,600

C. VHF Radios 4 @ \$500 ea \$2,000

Total \$8,600

Line 500 - Analysis

Salary time included in Line-100

A. Analytical hardware and software \$6,000

B. Report production costs \$1,000

Total \$7,000

B. Personnel qualifications

James Bodkin received a B.S. in Biology from California State University at Long Beach in 1976. He received an M.S. in Wildlife Biology from California Polytechnic University, San Luis Obispo in 1986. Since 1980 he has been employed by the U.S. Fish and Wildlife Service in Alaska and California. He worked on the California Sea Otter Project from 1980-1989. He is currently the Sea Otter Research Project Leader for the Alaska Fish and Wildlife Research Center. Jim has conducted sea otter research along the coast of North America between Attu, AK and Southern California, for nearly a decade.

Mark Udevitz received a B.S. degree in Wildlife Biology from Colorado State University in 1979, an M.S. degree in Wildlife Management from West Virginia University in 1982 and a Master of Statistics degree from North Carolina State University in 1986. He earned a Ph.D. in Biomathematics and Statistics in 1990 from North Carolina State University with a dissertation in the area of wildlife population estimation. He worked as a statistical consultant with the Southeastern Cooperative Wildlife and Fisheries Statistics Project from 1983-1989 and is currently working as the Statistician for the Mammals Branch of the Alaska Fish and Wildlife Research Center.

10. ANTICIPATED PRODUCTS

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It is anticipated that the results of this study will lead to the implementation of an accurate and precise sea otter census methodology in Prince William Sound. We anticipate results of this study to lead to the development of a standardized procedure that may be used throughout the North Pacific Ocean to evaluate sea otter abundance. We anticipate the results to be published in a refereed journal or as a monograph. This study will end with the development and implementation of a sea otter census protocol. For the purpose of the restoration process, continuation of the established protocols may be continued under a separate study plan.

Data storage:

Data will be managed and stored by sea otter project staff, under direction of the sea otter research project leader, at the Alaska Fish and Wildlife Research Center. Back-up copies of electronic data will be regularly maintained and original hard copies of data will be stored at a separate location.

Relationship to other FWS work:

The results of this study will potentially have broad applications within the Fish and Wildlife Service. Sea otters presently occur at three separate locations in the continuous US and several separate populations in Alaska as well as Canada and the Soviet Union. Current censuses of sea otter populations are not standardized and evaluation of sea otter populations are difficult. We anticipate the results of this study to have significant value to managers and scientists from both within Fish and Wildlife Service and other public agencies and private interests.

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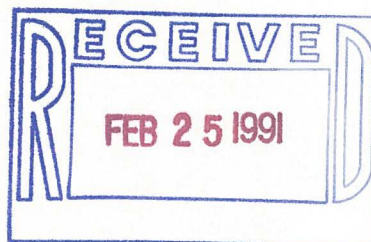
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PRE-SPILL AND POST-SPILL CONCENTRATIONS OF HYDROCARBONS
IN SEDIMENTS AND MUSSELS AT INTERTIDAL SITES
IN PRINCE WILLIAM SOUND

Recovery Monitoring Study Number _____

John F. Karinen and Malin M. Babcock
Principal Investigators

Auke Bay Laboratory
Alaska Fisheries Science Center
National Marine Fisheries Service, NOAA
P.O. Box 210155, Auke Bay, Alaska 99821



Lead Agency - NOAA, NMFS

Cost - \$83.9 K

Dates of Study Plan - 1 March 1991 to 29 February 1992

Signature Date

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John F. Karinen 21 Feb. 91

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INTRODUCTION

This proposal is for funding a second field sampling trip that will complement the Natural Resource Damage Assessment (NRDA) Coastal Habitat Study No. 2.

Hydrocarbon baseline information is available for 10 sites in Prince William Sound (PWS) prior to oil transport and for the first four years of oil shipment. This baseline for hydrocarbon levels in mussels, sediment, water, and fish was established from 1977 to 1981. All sites are located on low energy, low gradient beaches, often associated with eel grass. All sites have adjacent bands of mussels (Mytilus trossulus).

In 1989, we resampled 10 historically (1977-1981) established intertidal baseline sites in PWS in response to the *Exxon Valdez* oil spill. Additionally, 10 sites (6 in PWS and 4 on the Kenai Peninsula) were established along the spill trajectory before oiling, and sampled after oiling to measure the increase of hydrocarbon levels in sediments and mussels resulting from the spill (Figure 1). Sampling was continued in 1990. The first analyses of 1989 samples [sediments for hydrocarbons (HC)] are currently being conducted; preliminary analyses of the first run indicates that the March/April, 1989, sampling was successful in establishing pre-impact HC levels at the historically established and newly established sites.

Most of the sites established are low energy, fine-grained beaches at the head of embayments; some of them are expected to show significantly elevated levels of hydrocarbons in sediments and mussels and can be expected to retain petroleum hydrocarbons over longer periods of time than sediments from higher energy beaches or those beaches that have been treated.

This study has been funded under a NRDA Coastal Habitat Contract for sampling in 1989 and 1990. NOAA/Auke Bay Laboratory proposed 2 sampling trips for 1991 (see Table 1 for sampling frequency); however, funding under the NRDA process was reduced to 1 field trip.

Table 1. Field sampling frequency for NRDA Coastal Habitat Study sites in Prince William Sound.

YEAR/MONTH	MARCH	APRIL	MAY	JUNE	JULY	AUGUST
1989	X	X	X	X		X
1990		X		X		X
1991		X				X
(FUTURE) 1992-95				(X)		

Depuration and recovery (return to background levels) of sediments and mussels is probably influenced differently during two periods of time over the course of a year. The first, and probably the most important, is driven by severe weather experienced during the late fall, winter and early spring storms; the second, from April to early September, is characterized by relatively mild weather conditions conducive to enhanced microbial degradation and physiological processes. Sediment and mussel samples taken in August, 1991, will allow detection of changes over this second period and these changes (rates of recovery) can then be compared to the actual sampling times in 1990 and 1989 (see Table 1). One sampling trip is insufficient to rigorously determine rates of depuration/recovery just 2.5 years after the event.

Petroleum hydrocarbons in sediments and mussel tissues will not be considered injury unless it is demonstrated that the concentrations are causing biological damage. This study will provide linkage to several other studies that concern species in a higher trophic levels; i.e., hydrocarbons in mussels accumulate in birds, fish, shellfish, and mammals that feed on mussels and can affect survival, physiology and reproduction. Some HC concentrations found in mussels analyzed to date are equivalent to levels known to affect survival, reproduction, or behavior in molluscs in general, or the exposed mussels do not retain their utility to other organisms in the food web as a result of oil contamination.

Recovery to pre-spill levels of petroleum hydrocarbons in sediments and mussels is expected to take several years. Bioavailability of HC to flora and fauna is expected to decline, and recovery of some biota to pre-spill levels may be demonstrated in three to five years. Results will provide an estimate of recovery rates for the oiled sites, whether treated or untreated, and compared to reference sites, will allow prediction of time required for recovery.

OBJECTIVES

1. For all sites sampled during the NRDA process - to estimate the hydrocarbon concentrations in mussels and sediments such that the estimate is within 15% of the actual concentration 95% of the time when total aromatic concentrations are greater than 200 ng/g dry wt.
2. To compare petroleum hydrocarbon levels at all sites over all sampling periods. This second sampling trip will provide information on recovery of HC levels over a biologically active period (April - August) 2+ years after the accident.

METHODS

Sampling. Transect lines for sediment samples are thirty meters (m) in length and located parallel to the water line at -0.75 m to +0.75 m (depending on specific site). Sediment samples will be collected in triplicate at each site by compositing 10 cores (dia 3.2 cm x depth 1.25 cm) taken at random along the transect for each sample. Composite sediments will be placed in chemically clean jars, placed in an ice chest with artificial ice and transported. These will be frozen within 2-3 hours of collection. One blank sample will be taken at each site.

Mussel transects are located in mussel bands, parallel to the water line, usually just up (~+1 m tide level) from the sediment transects. Triplicate mussel samples will be collected by taking approximately 30 2-5 cm. mussels (enough to produce ≥ 10 gm tissue) at random along the 30-meter transect. Samples in 16 oz. jars will be cooled, transported and frozen in the same manner as the sediment samples.

Quality assurance and quality control (QA/QC) plans. In the matter of data collection and analysis, sample collection (with labelling), and chain-of-custody procedures, we will adhere to guidelines as developed by the NRDA Hydrocarbon Technical Committee and implemented by NOAA/Auke Bay Laboratory. The QA/QC for analytical chemistry, and Procedures for sample collection/chain-of-custody for NOAA/Auke Bay Laboratory are attached.

DATA ANALYSES

Random sample and subsample collection up to the analysis procedure should assure that hydrocarbons present in the sample represent the average concentration at each site. "Hot spots" of hydrocarbon concentration over the 30 meter transects should be cancelled out by this procedure.

All hydrocarbon level data among sites will be analyzed by ANOVA. For comparison over time, a repeated ANOVA will be used. Further multiple (Scheffe') or paired (Tukey) methods may be used to test differences at selected sites and will be tested at $p=.05$.

Sources of the hydrocarbons over time (natural or anthropogenic) will be determined by examining the relative composition of various components of aromatics and alkanes, i.e. odd/even ratios of alkanes, pristane/phytane ratio, 1 and 2 ring aromatic abundance versus 4 and 5 ring compounds, relative numbers of substituted aromatic compounds versus parent compounds, comparison of hydrocarbon patterns with those of Prudhoe Bay

crude oil, etc.

All data will be presented in tabular and graphic forms, and selected data will be mapped.

SCHEDULES AND PLANNING

Data and report submission schedule. This proposal is for sediment and mussel sampling during the lowest tide series in August 1991. Covering all 16 sites during this short period (7 days), requires transportation of the field team by helicopter. The team will probably be based out of Valdez, Alaska.

Data compilation, analyses timetable and report writing will be accomplished according to a schedule to be set by the Restoration Planning Work Group and the Trustee Council. Issuing of reports will necessarily be controlled by completion of chemical analyses of the samples collected. Interim status reports will be written as required.

Sample and data archives. Study Plans (with revisions), QA/QC plans, data sets, log books, etc. will be stored under secured conditions as prescribed by the Trustees and implemented by NOAA.

Management plan. This one field trip will be conducted by the two Principal Investigators, John Karinen and Malin Babcock, plus 1 field party member (as yet, unidentified).

Logistics. See first paragraph under this heading.

PERSONAL QUALIFICATIONS

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Education

1957 B.S., Biological and Physical Science, Black Hills State College, Spearfish, South Dakota
1958 B.S., Education, Black Hills State College, Spearfish, South Dakota
1965 M.S., Biological Oceanography, Oregon State University, Corvallis, Oregon
1965-91 Various graduate and training courses: OSU, University of Alaska, Federal government

Professional Positions

1989-present: Habitat Investigations, ABL - Fishery Research Biologist; Principal Investigator, *Exxon Valdez* Natural Resource Damage Assessment Study "Pre-Spill and Post-Spill Concentrations of Hydrocarbons in Sediments and Mussels at Intertidal Sites in Prince William Sound." General consultant in patterns and implication of hydrocarbons in sediments and fauna. Member, Technical Advisory Group for the Alyeska Ballast Effluent Facility.

1985-1989: Marine Investigations, ABL - Supervisory Oceanographer, Groundfish: Included resource assessment and prediction of stock abundance, distribution and recruitment with emphasis on demersal fish in eastern Gulf of Alaska.

1972-1985: Habitat Investigations, ABL - Task Leader, Oil effects studies: Laboratory and field-oriented oil effects studies (established historical baseline sites for hydrocarbon levels in sediments and biota in Prince William Sound); supervised staff of 10 scientists; general consultant - environmental impacts (mining, oil and marina development).

Selected Publications

Karinen, John F. 1988. Sublethal effects of petroleum on biota. Pp. 294-328 in Shaw, D. G. and M. J. Hameedi (eds.), *Environmental Studies in Port Valdez, Alaska: A Basis for Management*. Lecture Notes on Coastal and Estuarine Studies 24. Springer-Verlag. New York.

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Karinen, J. F. 1980. Petroleum in the deep sea environment: Potential for damage to biota. Environ. Int. 3(2):135-144.

PERSONAL QUALIFICATIONS

MALIN MARIE BABCOCK

Professional Address NOAA, National Marine Fisheries Service
Auke Bay Laboratory (ABL)
P. O. Box 210155
Auke Bay, Alaska 99821-0155
(907) 789-6018

Education

1962 B.S. Zoology, Oregon State University, Corvallis, Oregon
1962-1964 Graduate course work in Zoology, Oceanography Oregon State University, Corvallis, Oregon
1969 M.S. Zoology (Fisheries), University of Alaska, Fairbanks (UAF)
1969-Present Continuing education in professional field, supervision, management, personal growth and writing areas through workshops, short courses and classes.

PROFESSIONAL HISTORY:

1987 - Present: Habitat Investigations, ABL - Task Leader and Supervisory Fishery Research Biologist: conduct and supervise research on critical habitat and physiological requirements in juvenile king crabs. Co-Principal Investigator (P.I.), Exxon Valdez Natural Resource Damage Assessment (NRDA) Study "Pre-Spill and Post-Spill Concentrations of Hydrocarbons in Sediments and Mussels at Intertidal Sites in Prince William Sound." One of ABL staff scientists to consult with ADF&G biologists in establishing experimental designs and study plans for there many NRDA projects. Supervise staff whose responsibilities include NRDA database management, QA/QC for sample collection and handling, and P.I.'s on other NRDA studies.

1973 - 1987. Habitat Investigations, ABL - Fishery Research Biologist: Research during this period was mainly laboratory oriented and concerned effects of hydrocarbon exposures to aquatic organisms. Assisted (1979 to 1981) in establishing base line hydrocarbons levels in sediments and selected organisms of subtidal and intertidal areas of Prince William Sound.

Selected Publications

Thomas, Robert E., Stanley D. Rice, Malin M. Babcock, and Adam Moles. 1989. Differences in hydrocarbon uptake and mixed function oxidase activity between juvenile and spawning adult

coho salmon (*Oncorhynchus kisutch*) exposed to Cook Inlet crude oil. *Comp. Biochem. Physiol.* 93C(1):155-159.

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Babcock, M. M. 1985. Morphology of olfactory epithelium of pink salmon, *Oncorhynchus gorbuscha*, and changes following exposure to benzene: A scanning electron microscopy study. Pp. 259-267 in Gray, J. S. and M. E. Christiansen (eds.), *Marine Biology of Polar Regions and Effects of Stress on Marine Organisms*. Proceedings of the 18th European Marine Biology Symposium, University of Oslo, Norway, 14-20 August 1983. John Wiley & Sons. Chichester, England.

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BUDGET

<u>Line Item</u>	<u>Amount (Thousands)</u>
Labor	8.0
Travel	6.0
Contracts: Helicopter	13.5
Sample Analysis*	55.9
Supplies & Equipment	<u>0.5</u>
TOTAL	83.9

*16 sites x 2 (sediment, mussels) x 3 (triplicate) = 96 samples
96 x \$582 (current analytical cost, ABL) = \$55,872.00

SELECTED LITERATURE

Connell, Joseph H. 1970. A predator-prey system in the marine intertidal region. 1. *Balanus glandula* and several predatory species of *Thais*. Ecol. Monog. 40:49-78.

Gundlach, Erich R., Paul D. Boehm, Michel Marchand, Ronald M. Atlas, David M. Ward, and Douglas Wolfe. 1983. The fate of Amoco Cadiz oil. Science 221:122-129.

Karinen, John F., L. Scott Ramos, Patty G. Prohaska, and William D. MacLeod, Jr. In Preparation. Hydrocarbon distribution in the marine environment of Port Valdez and Prince William Sound, Alaska.

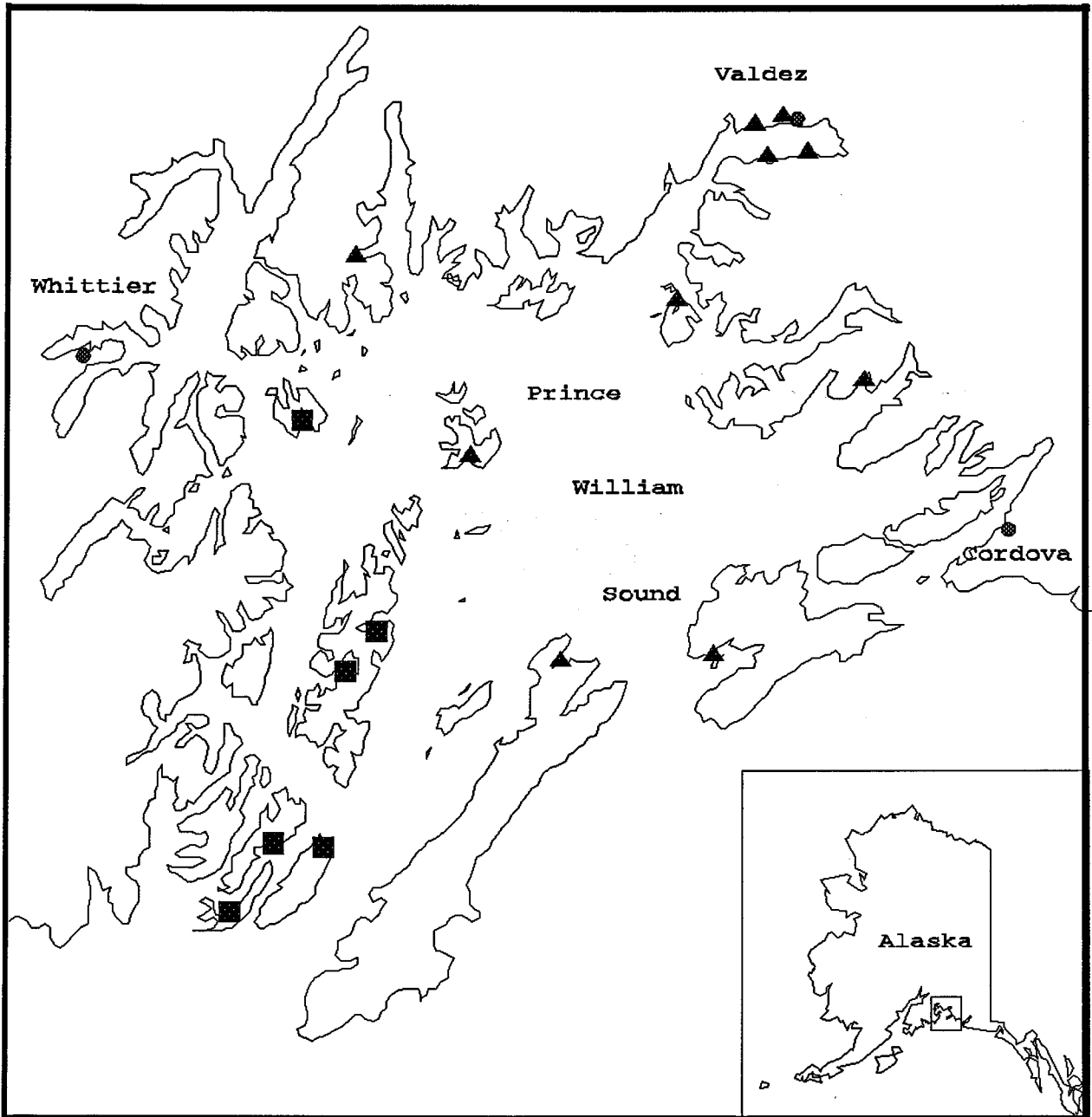
Karinen, John F. 1988. Sublethal effects of petroleum on biota. Pp. 294-328 in Shaw. D. G. and M. J. Hameedi (eds.), Environmental Studies in Port Valdez, Alaska: A Basis for Management. Lecture Notes on Coastal and Estuarine Studies 24. Springer-Verlag. New York.

Krahn, M. M., C. A. Wigren, R. W. Pearch, L. K. Morre, R. G. Bogar, W. D. MacLeod, Jr., S. Chan, and D. W. Brown. 1988. Standard analytical procedures of the NOAA National Analytical Facility, 1988, New HPLC cleanup and revised extraction procedures for organic contaminants. NOAA Tech. Memor. NMFS F/NWC-153. 52 pp.

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Figure 1. Intertidal baseline sampling sites.

▲ = historical sites ■ = established in 1989.



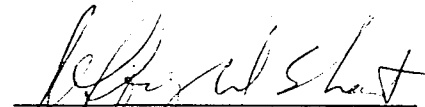
State/Federal Natural Resource Damage Assessment
for the

T/V Exxon Valdez Oil Spill

Analytical Chemistry Quality Assurance Plan


Auke Bay Laboratory
Alaska Fisheries Science Center
National Marine Fisheries Service, NOAA
P. O. Box 210155, Auke Bay, Alaska 99821

Submitted:


Jeffrey W. Short
Research Chemist
Auke Bay Laboratory

Dated: November 16, 1989

Accepted:


Dr. Carol-Ann Manen
Analytical Chemistry
Group Representative

Dated: 11/30/89

I. Introduction

This Plan describes the procedures that will be used at the Auke Bay Laboratory (ABL) for the analysis of hydrocarbons in samples collected for the State/Federal damage assessment of the T/V Exxon Valdez oil spill in Prince William Sound, Alaska. These samples, consisting of water, sediment, and biological tissues, are collected by other participants in the damage assessment process. Collected samples are archived by the Analytical Chemistry Group (ACG), and the ACG subsequently selects samples to be sent to participating analytical laboratories for chemical analysis. Laboratory participation is contingent on demonstration of analytical capability, and approval of a Quality Assurance Plan (QAP) by the ACG. This document is the QAP for ABL, and describes the standard operating procedures (SOP's) that will be used at ABL associated with these hydrocarbon analyses, the quality control (QC) measures to be used, the quality assurance (QA) criteria to be used, and the data deliverables that will result from these analyses.

II. Standard Operating Procedures

A. Chain of Custody

Project samples (PS's) will be supplied to ABL by the ACG, together with sufficient information on the PS's to allow interpretation of analytical results, a chain of custody form for each sample supplied, and an indication of the hydrocarbon contamination level expected in each sample. Receipt of PS's will be acknowledged by Mr. Tony Chan of ABL on the chain of custody form, who will store the received PS's in a locked freezer at ABL and will file the chain of custody form in a locked cabinet at ABL. The sample identification number (SIN) of each sample provided on the chain of custody form will be recorded in a bound laboratory research notebook by Mr. Chan. Each time material is transferred from one container to another during sample processing, the transfer will be witnessed by another ABL staff member, and both the transferrer and the witness will date and sign an entry in the laboratory notebook describing the transfer. The notebook entry will include, at a minimum, the SIN of the sample being transferred, the name of the transferrer, and a reference to the analytical method justifying the transfer. Unused material of each PS will remain in the locked freezer. Each sample container used in the analytical process will be marked with the SIN, and the validity of the SIN's on the sample containers will be acknowledged by the witness of the transfer in the transferrers laboratory notebook. When a sample container is transferred among ABL staff members, the transfer will be noted in the laboratory notebooks of the participating members, who will each witness the transfer by signing and dating all the laboratory notebooks involved in the transfer. These laboratory notebooks will act as continuations of the chain of custody form for each PS while at ABL.

B. Sample Preparation

Project samples will be extracted and prepared for gas chromatography using procedures described by Krahn et al. (1988). In particular, preparation of sediment samples will follow sections 2 and 4 of Krahn et al. (1988), except that the portions of this procedure pertaining to analytes other than aromatic hydrocarbons will be ignored. Similarly, tissue samples will follow sections 3 and 4 of Krahn et al. (1988). Methylene chloride extracts of water samples will be prepared in the same way as sediment samples, except step B.2, and steps B.7 through B.21, inclusive, of section 2 will be omitted. Internal and calibration standards will be identical with those described in section 1 of Krahn et al. (1988). The sequence of steps summarizing sample preparation described by Krahn et al. (1988) is initial extraction (sediment and tissue only), extract concentration and silica gel chromatography clean up, extract reconcentration and HPLC (gel permeation) clean up, and final extract reconcentration. The final product of this sample preparation procedure is about 250 μ l hexane extract, which is split equally among two GC sample vials. One of the GC vials is used for immediate GC/FID analysis, and the other is stored in case the analytical results of the analysed vial are not acceptable.

C. GC Analysis

Final sample extracts will be analysed by GC/FID using the method described in section 12 of MacLeod et al. (1985). Amounts of aromatic hydrocarbons contained in National Institute of Standards and Technology (NIST) Standard Reference Material (SRM) 1491 will be quantitatively determined, and will hereafter be denoted by the term "analyte". Samples containing sufficient amounts of these aromatic hydrocarbon analytes to be detectable by GC/MS using selected ion monitoring will be so analysed to confirm analyte identity, after consultation for approval with Dr. Carol-Ann Manen (ACG chair) and the principal investigator of the project that generated the PS. The GC/MS will be tuned using PFTBA, and will monitor the suspected parent ion and at least one confirming ion for each measured analyte. GC operating conditions and column will be the same as those used for the GC/FID.

D. Dry Weight Determination

The ratio of wet weight to dry weight will be determined for each sediment and tissue PS using the method described in section 9 of MacLeod et al. (1985).

III. Quality Control

A. Sample Batches and Strings

Project samples will be analysed in batches which will include 12 PS's per batch. The PS's of a batch will be of the same project and matrix. A reagent blank sample, a reagent blank sample spiked

with the SRM 1491 calibration standard, and two reference material samples (supplied by NIST) will be included with each batch of PS's and processed identically with the PS's. Each batch of samples analysed by GC/FID or by GC/MS will be analysed in a sample string consisting of the batch samples plus additional samples in the following sequence:

<u>Sequence Number</u>	<u>Sample Identity</u>
1	Hexane Blank
2	Calibration Standard #1
3	Calibration Standard #2
4	Calibration Standard #3
5	Hexane Blank
6 thru 11	Project Samples
12	Hexane Blank
13	Calibration Standard #2
14	Reagent Blank
15	Reference Material
16 thru 21	Project Samples
22	Spiked Reagent Blank
23	Reference Material
24	Calibration Standard #2

B. Internal Standards

Analyte internal standard (A/IS) is added to each PS, reagent blank, spiked reagent blank, and reference material sample at the beginning of the sample preparation sequence, consisting of 5.01 μg each naphthalene- d_8 , acenaphthene- d_{10} , benzo[a]pyrene- d_{12} , and perylene- d_{12} . An independent internal standard consisting of 5.00 μg phenanthrene- d_{10} (HPLC/IS) is added to each reconcentrated extract just prior to HPLC clean up to determine the recovery of the A/IS's. Another independent internal standard consisting of 2.40 μg hexamethylbenzene (GC/IS) is added to each final reconcentrated extract to determine recovery of the HPLC/IS. The A/IS and the GC/IS are supplied by NIST; the HPLC/IS will be prepared at ABL.

C. Calibration Standards

The calibration standards are a mixture of SRM 1491, and the internal standards A/IS, HPLC/IS, and GC/IS. The amounts of internal standards present in the calibration standards are the same as in the PS's, reagent blanks, reagent blanks with SRM 1491 spikes, and reference material samples. The volume percent of SRM 1491 in calibration standards 1, 2, and 3 is 5.08%, 20.3%, and 81.3%, respectively. The remaining volume of the calibration standards is either internal standard or hexane.

D. Performance Monitoring

The following will be evaluated for each sample string analysed, and will form the basis for determining the acceptability of data resulting from the analysis.

1. Instrument Precision

Instrument precision will be determined by calculating the mean detector response for each analyte in the three calibration standard #2's, and then calculating the detector response deviation from this mean for each analyte in each of the three calibrations standard #2's. The results of repeated analysis of this calibration standard among sample strings will be tracked using a control chart, and the comparison of the calibration standard results for a string with the control chart results will be a determinant of data acceptability.

2. Method Precision

The evaluation of method precision for each analyte will be calculated as the variance of the results of reference material analysis among strings. The results of repeated analysis of the reference material will be tracked using a control chart, and the comparison of the reference material results for a string with the control chart results will be a determinant of data acceptability.

3. Calibration Curve Linearity

The linearity of the GC/FID or GC/MS calibration curve for each analyte will be determined by comparing the analyte response factor in each of the calibration standards 1, 2, and 3. The analyte response factor is the ratio of the amount of analyte present in the calibration standard and the magnitude of the detector response.

4. Reagent Spike Recovery

Reagent spike recovery will be calculated as the ratio of analyte found in the reagent spike sample (less that found in the reagent blank), and the known amount initially added in the spike.

5. HPLC Internal Standard Recovery

HPLC/IS recovery will be calculated as the ratio of the amount of HPLC/IS found in a sample based on the GC/IS, and the amount of HPLC/IS initially added to the sample.

6. Aromatic Hydrocarbon Internal Standard Recovery

A/IS recovery will be calculated as the ratio of the amount of A/IS found in a sample based on the HPLC/IS, and the amount of A/IS initially added to the sample.

7. Reagent Blank Contamination

Reagent blanks will be examined for detector response at retention times corresponding with analyte retention times, to verify the absence of contamination during sample preparation.

8. Hexane Blank Contamination

Hexane blanks will be examined for detector response at retention times corresponding with analyte retention times, to verify the absence of sample carry-over during GC analysis.

IV. Quality Assurance

Following are the criteria that will be used to determine the data acceptability for each sample string analysed, and recourse for sample strings yielding unacceptable data.

A. Acceptance Criteria

Data for each sample string will be considered acceptable if all of the following criteria are met:

1. Instrument Precision: Detector response for each analyte in each calibration standard #2 is within $\pm 10\%$ of the mean detector response calculated from all three calibration standard #2's. The deviation of these means among strings is $< 20\%$ to insure detector sensitivity.

2. Method Precision: The concentration of each analyte found in the reference material is $< \pm 35\%$ among strings, and the average deviation for all analytes is $< 30\%$ among strings.

3. Calibration Curve Linearity: Response factors for each analyte in calibration standards 1, 2, and 3 analysed at the beginning of each sample string are within $\pm 10\%$ of the mean response factor calculated from these calibration standards.

4. Reagent Spike Recovery: The ratio of analyte detector response and A/IS detector response in the spiked reagent blank is within $\pm 15\%$ of the average of this same ratio in the three calibration standard #2.

5. HPLC/IS Recovery: $> 70\%$

6. A/IS Recovery: $> 70\%$

7. Reagent Blank Contamination: No detector response significantly different than baseline noise.

8. Hexane Blank Contamination: No detector response significantly different than baseline noise.

B. Reanalysis of Unacceptable Batches

Project samples that contain analytes at concentrations above the linear range spanned by the calibration standards will be diluted and analysed at the end of the sample string. Otherwise, samples associated with sample strings that do not satisfy the quality assurance criteria (IV.A above) will be reanalysed after the fault is identified and corrected. If necessary, these samples will be re-processed to provide sufficient extract for re-analysis.

V. Data Processing and Verification

Resident software on the GC/FID or GC/MS systems will be used to translate detector response for each analyte into matrix concentrations for that analyte. Computer programs will be written for this purpose, and will be verified by comparison with hand calculated results prior to program implementation.

VI. Data Deliverables

Data deliverables will consist of computer files containing all data associated with each sample analysed. These files will contain the concentration of each analyte found, the data associated with sample collection, the QC data collected for the batch containing that sample, and the GC system stability and detector linearity verification data for the batch. These data will be stored together in a data base management system such as RBASE. A written report summarizing the results and QC data will be prepared by the principal investigator. Included in the report will be quantitative estimates of the precision and accuracy of the results.

VI. Detection Limit Evaluation

Method detection limits for each analyte and matrix will be determined at least twice annually using methods specified in Appendix B, 40 CFR Part 136.

VII. Technical System and Performance Audits

The laboratory and staff analysing these samples will be available for audit without prior notice by qualified representatives of any of the Trustee agencies, with the understanding that the auditors will present a written description of the specific audit goals and objectives at the commencement of the audit. It is further understood that ABL will participate in a minimum of three laboratory intercomparison exercises yearly which will be coordinated by NIST and will involve the blind analysis of gravimetrically

prepared materials, extracts of environmental matrices (tissue, sediment, and water) or the matrices themselves.

VIII. Literature Cited

Krahn, M. M., Wigren, C. A., Pearce, R. W., Moore, L. K., Bogar, R. G., MacLeod, W. D. Jr., Chan, S. L., Brown, D. W. 1988. "Standard Analytical Procedures of the NOAA National Analytical Facility, 1988. New HPLC Cleanup and Revised Extraction Procedures for Organic Contaminants." NOAA Technical Memorandum NMFS F/NWC-153, U. S. Department of Commerce, National Oceanic and Atmospheric Administration, National Marine Fisheries Service.

MacLeod, W. D. Jr., D. W. Brown, A. J. Friedman, D. G. Burrows, O. Maynes, R. W. Pearce, C. A. Wigren, R. G. Bogar. 1985. "Standard Analytical Procedures of the NOAA National Analytical Facility, 1985-1986. Extractable Toxic Organic Compounds." NOAA Technical Memorandum NMFS F/NWC-92, U. S. Department of Commerce, National Oceanic and Atmospheric Administration, National Marine Fisheries Service.

Damage Assessment Custody Forms and Data Entry
NMFS Auke Bay Laboratory, Box 210155, Auke Bay AK 99821 February 14, 1991

CUSTODY FORMS

Enclosed are new custody forms to be used for all DAD samples sent to Auke Bay Lab for analysis in 1991. These forms differ very little from those used in 1990, but please read these instructions carefully whether or not you have used similar custody forms before. Note that each line on the form has an **ASSIGNED NUMBER**, in the first field. **This number must be written on the label of the sample that is entered on that line of the custody form!** No samples will be accepted for hydrocarbon analysis by the Auke Bay Laboratory unless they are accompanied by these pre-numbered custody forms and bear the assigned numbers on their labels.

These custody forms serve two functions: 1) to satisfy the legal requirement of documenting the chain of custody of samples, and 2) to provide the data we need for the DAD database. You may elect to send the data electronically, in which case the custody form would only need enough data filled in to satisfy legal requirements. Even if the entire data set is sent electronically, the samples must be accompanied by a custody form that includes date collected and matrix for each sample, and has all parts completed under "Chain of Custody" on the bottom half of the form. See the Electronic Data Input and Custody Form Data Input sections below. We prefer that data be sent to us electronically, but if you cannot do so then complete *all* fields on the custody form and we will have what we need. Each sample should be one line on the custody sheet or electronic file. Do not list more than one sample on one line of the custody form.

These chain-of-custody forms are accountable forms; you are responsible for them. If you pass any on to other users, let us know who they go to. Return all unused or voided forms to us by the end of 1991. New forms will be issued for 1992.

SHIPMENT OF SAMPLES

Avoid costly damage to your valuable samples:

- **Always** arrange with Sid Korn (907) 789-6021 *before* shipping samples.
- **Always** ship samples in a cooler or insulated container. Inadequately insulated samples thaw in transit, which seriously lowers sample quality and priority for analysis.
- **Do not** use commuter airlines or courier services to ship samples. They are too slow, and we have had even well-packed samples thaw when shipped this way.
- **Do** hand carry samples as excess baggage to Juneau if possible or hand carry samples to Anchorage, and ship them by Alaska Airlines as frozen air freight. Monitor the shipment, to see that it is held in the freezer until shipment and to determine which flight it is on. When shipping from Kodiak, Valdez, Kenai, etc. *please follow this procedure and ship from Anchorage.*

CUSTODY FORM DATA INPUT

The following should answer most questions that arise regarding input of sample data on custody forms.

VAL-91- Leave blank. (This is a batch number assigned after the samples reach ABL.)

Project _____ Your official NRDA project title, e.g. Air/Water 2, Fish/Shellfish 5.

Assigned Sample # This number must appear on the sample label. Note that the first 4 digits are the same as the form serial number, and only the final two digits are printed on most sample lines. *Put the whole 6-digit number on each sample jar.* If you have less than 50 samples in one batch, do not use the rest of the numbers; if you have more than 50 samples, use more than one custody form.

Collector's Sample Code Optional. If you use a numbering scheme other than the pre-assigned numbers, you may enter your sample numbers here and they will be included in the data base for your convenience.

Date Collected Date each sample was collected from the field.

Matrix The material sampled, usually one of the following: sediment, tissue, water, or sample blank, see code list. NOTE: If the matrix is tissue, specify species as accurately as possible (see code list) and sub-matrix (e.g. liver, bile, whole body; see code list).

Location Collected Specific geographic location name, (e.g. Snug Harbor; see code list).

Latitude/Longitude Degrees, minutes, and seconds, for your specific sample location.

Depth In meters, for underwater samples only.

Method Sample collection method; see code list.

- If there is no appropriate entry listed on a code list, fill in the actual data (genus & species, submatrix, location, method) and we will assign a new code when entering the data in the data base.
- If you have more than one sample from the same animal (e.g. both muscle and bile samples from one fish) be sure to indicate which samples came from the same animal, in any convenient manner.

Linda

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PRINCE WILLIAM SOUND PINK SALMON

ESCAPEMENT ENUMERATION

Project Number:

Principal Investigator: Samuel Sharr

Lead Agency: Alaska Department of Fish and Game

Cost of Proposal: \$230,000

Project Dates: July 1, 1991 - June 30, 1992

Principal Investigator: _____

Organization Leader: _____

Finance Officer: _____

INTRODUCTION

Wild stock production of pink salmon in PWS has ranged from 10 to 15 million fish in recent years. Chum salmon returns have ranged from .8 to 1.5 million fish. Much of the spawning for pink and chum salmon (up to 75% in some years) occurs in intertidal areas. Intertidal spawning areas are susceptible to marine contaminants and there is strong evidence the March 24, 1989, Exxon Valdez Oil Spill (EVOS) adversely affected spawning success and early marine survival in Prince William Sound. Salmon stocks impacted by the Exxon Valdez Oil Spill (EVOS) are also heavily exploited in commercial, sport, and subsistence fisheries. These stocks can most effectively be restored through stock specific management practices designed to reduce exploitation on impacted stocks. The stocks in areas heavily impacted by the EVOS are present in fisheries dominated by hatchery and wild stocks from unaffected areas of the Sound. The management of this mixed stock fishery has historically been based on maintaining good temporal and spatial distribution of spawning escapement for groups of stocks in eight major fishing districts. The success of this management strategy was predicated upon the effectiveness of the aerial survey program as an inseason escapement estimation tool. Restoration premised on stock specific management of the commercial fishery for reduced exploitation of impacted stocks will require even more accurate inseason escapement estimates for impacted and unimpacted wild stocks.

This project is designed to provide accurate, real time, escapement estimates for salmon stocks of Prince William Sound. Accurate inseason escapement estimates will enable fisheries managers to identify shortfalls in the numbers of spawning fish in impacted streams and impose fisheries restriction to reduce harvest rates on those stocks. The manager will also be able to identify any excesses in escapement and direct very localized fishing effort to harvest surplus fish. Post season analyses of the escapement enumeration project together with results from the proposed Coded-Wire Tagging project will provide stock specific estimates of total return and enable managers to assess the effectiveness of their stock specific management strategies.

In the absence of improved stock specific management capabilities afforded by this project, salmon stocks in western PWS which have already been stressed and depleted by the oil impacts will potentially be over exploited in the commercial, sport and subsistence fisheries. Population levels may be reduced below those needed for rapid recovery and in some instances may result in virtual elimination of impacted stocks.

The foundations for this project were firmly established during the damage assessment process in Natural Resources Damage Assessment (NRDA) Fish/Shellfish (F/S) Study #1. Extent of oiling in intertidal spawning areas was documented and escapement enumeration procedures were developed and perfected. In 1989 a total of 411 streams were surveyed for the presence of oil in intertidal spawning areas and 138 streams from among the 218 in the historic aerial survey program were included in a ground census of pink and chum salmon escapements. In 1990 the oil survey was limited to the 138 streams in the escapement censusing portion of the

project. Mussel samples for hydrocarbon analysis were collected in the intertidal mouths of the 138 streams in the ground censusing program in both 1989 and 1990. Total area of intertidal spawning habitat was estimated for each of the 138 streams and the area of upstream spawning habitat was estimated for 100 of the 138 streams. Total spawning escapement at four streams was estimated through weirs and stream residence time (stream life) estimates were made for pink salmon in 22 streams in 1990. Tissue samples for hydrocarbon analysis were collected from spawning adult pink salmon in 12 oiled and 10 unoiled streams in the ground survey program.

This program is designed for stock specific restoration and will emphasis more detailed and intensive data collection on fewer streams in the oil impacted areas of western PWS. Weirs will be installed on four streams weired in 1990 plus three additional streams. Six of these streams were in the wild stock coded wire tagging study (formerly NRDA F/S Study #3). Adults returns will be enumerated and be sampled for coded wire tags applied during the 1990 field season. Ground surveys and stream life studies will be continued at each weired stream and approximately 21 additional streams. Oil surveys as well as mussel and adult salmon tissue sampling will continue on all surveyed streams for the duration of the project.

The results of the study will provide estimates of average stream life for pink and chum salmon in PWS, will calculate coefficients to adjust for bias in aerial survey counts based on comparisons with accurate counts through weirs, and will use the stream life estimates and calibration coefficients to make accurate escapement estimates for the current year and all prior years for streams included in the ADF&G aerial survey program. Historic aerial survey data will be used to build timing curves and develop escapement goals for oil impacted stocks. Management strategies for oiled stocks in 1991 and succeeding years will be based on comparisons of these timing curves and escapement goals with inseason escapement data. In addition, results of this study will provide estimates of post oil spill spawning distribution within stream zones and among streams; will estimate total available intertidal and upstream spawning habitat for each stream; will provide marine survival estimates for six wild stocks of pink salmon based on coded wire tagging and recovery; will document physical presence or absence of oil in intertidal salmon spawning and rearing habitat and presence or absence of oil in the tissues of mussels and salmon which rear or live there; and will provide an atlas of aerial photographs and detailed maps for important spawning sites.

OBJECTIVES

- A. Enumerate the total intertidal and upstream pink and chum salmon escapement through weirs installed on seven moderately large streams which are representative of streams in the aerial and ground escapement survey programs.
- B. Estimate the number of spawning salmon, by species, within standardized

intertidal and upstream zones for 27 streams in PWS.

- C. Estimate the accuracy of aerial counts for the 218 aerial index streams by comparison of paired ground and aerial counts from the same streams on the same or adjacent survey dates and by comparison of aerial, ground, and weir counts on seven stream.
- D. Estimate average stream life of pink and chum salmon in at least 27 streams in PWS using a variety of techniques.
- E. Estimate 1961 through 1988 pink and chum salmon escapements to the 218 aerial index streams using the average observed error in the aerial survey method and stream life data from 1989, 1990, and 1991.
- F. Develop escapement goals and timing curves for all stocks in the ADF&G aerial survey program for inseason stock specific management of the commercial fisheries.
- G. Enumerate adult returns in streams where coded wire tags were applied to wild pink salmon stocks and assist in the spawning ground sampling for tag recovery.
- H. Produce a catalog of aerial photographs and detailed spawner distribution maps for the more important pink and chum salmon streams of Prince William Sound for use in designing sampling transects in the egg deposition and pre-emergent fry studies.
- I. Continue to document presence or absence of oil on intertidal habitat used by spawning salmon through visual observation, aerial photography, and hydrocarbon analysis of tissue samples from intertidal mussels at stream mouth.
- J. Continue to document the physical extent of oil distribution on intertidal spawning areas.
- K. Document presence or absence of hydrocarbons from the EVOS in tissues of adult salmon returning from fry outmigrations which occurred in 1989 and subsequent years in oiled and unoiled areas.

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 or identical to those used in NRDA F/S Study #1. These procedures have been thoroughly reviewed by the NRDA peer review process and approved by the Management Team.

The technology and methodology for escapement enumeration using systematic aerial and ground survey programs, as well as weir projects are well established and have a long history of success in Alaska. The historic aerial and ground survey data base for Prince William Sound is one of the best in the world. This data base provides the principal inseason management tool for wild pink and chum salmon stocks and will be critical to stock specific restoration efforts. The existing NRDA projects greatly enlarged the scope of the pre-spill escapement enumeration projects. The proposed salmon escapement enumeration projects for restoration will improve fisheries management and are a logical extension to the existing management programs and the NRDA process.

Aerial Surveys

Aerial survey estimates of pink and chum salmon numbers in 209 index streams will be flown by personnel from ADF&G Division of Commercial Fisheries as they have been since 1961 (Figure 1). Eight additional streams in oiled areas were incorporated into the program in 1989. Surveys are flown weekly from mid-June to mid-September each year. Counts of live salmon by species will be recorded for the bay at the terminus of each stream, the mouth of each stream, and within the stream (Pirtle, 1977). In 1990 the frequency of survey flights almost doubled and in most weeks there was at least two observations per stream. This increased survey frequency will continue in 1991 if funding from the local aquaculture association is sustained.

Total Enumeration Studies

Weirs for total escapement enumeration will be installed on seven streams in 1991 (Figure 2). The four streams weired in 1990 as part of NRDA Study #1 will among those weired in 1991. The six streams in the coded-wire tagging project for wild stocks of pink salmon (NRDA F/S Study 3) will also be a subset of the weired systems. The weirs will be installed at or as near as possible to the 1.8 meter tide level or the lower level of intertidal spawning. Weir crews will record daily fish passage through the weir.

Ground Surveys of Escapements

The 28 streams (Figure 2) to be surveyed will be selected according to the following criteria:

1. stream is included in the ADF&G aerial survey program;
2. stream is included in the pink and chum salmon egg deposition and pre-emergent fry project (NRDA F/S Study 2);
3. stream is included in the CWT project for wild stocks of pink salmon (NRDA F/S Study 3);

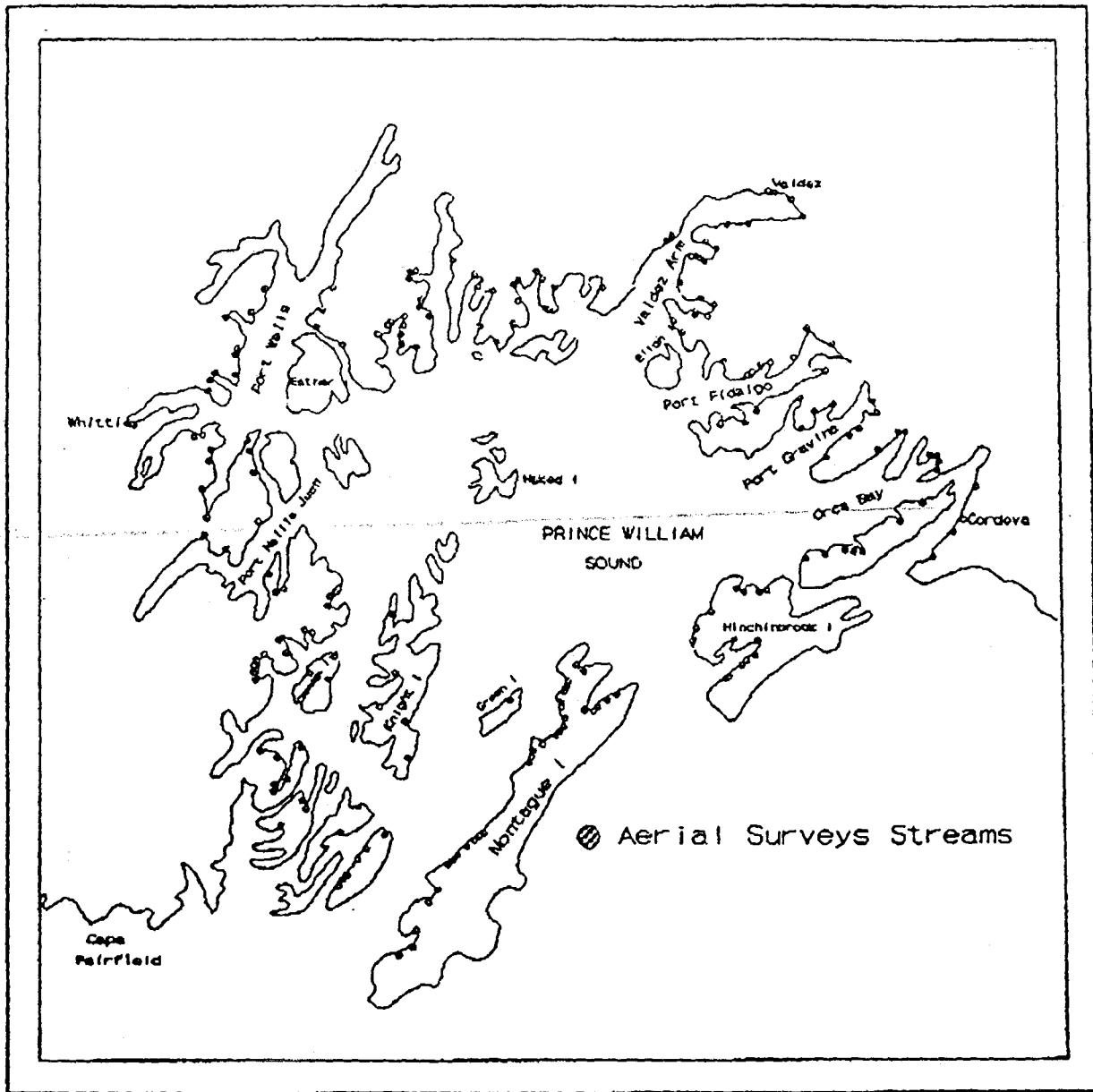


Figure 1. Streams included in the aerial survey programs for estimating pink and chum salmon escapement to Prince William Sound.

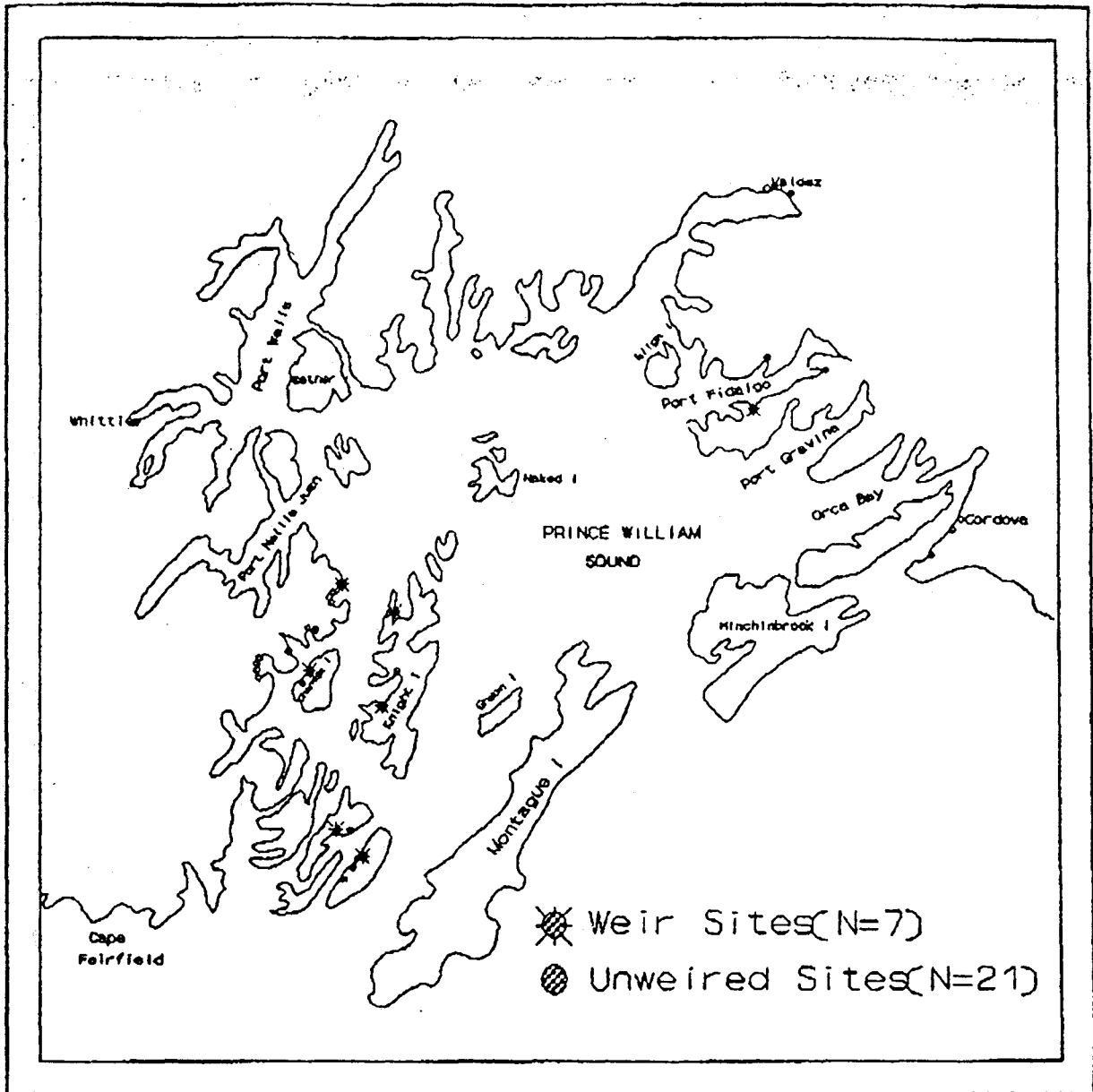


Figure 2. Streams proposed for the weir, ground survey, and stream life studies in 1991.

4. stream has been included in stream life studies conducted by this project in 1989 and 1990;
5. stream was enumerated in prior spawning ground foot survey programs and;
6. streams included will be from both oiled and unoled areas.

Maps of all streams in the program prepared from aerial photographs and modified and corrected during NRDA F/S Study #1 in 1989 and 1990 will be used and updated during the 1991 field season.

The pre-season survey to mark tide zones will be conducted in June, prior to the return of the pink and chum salmon. The location of tide levels 1.8, 2.4, 3.0, and 3.7 m above mean low water will be measured from sea level using a surveyors's level and stadia rod. Sea level at each site will be referenced to mean low water with site specific, computer generated tide tables which predict tides at five minute intervals. Tide zone boundaries will be delineated with color coded steel stakes. The linear length of the stream within each intertidal zone will be measured with a surveyors chain or range finder. The linear length of the stream in the upstream zone will be measured similarly on short streams and estimated from accurately scaled aerial photos on long streams. The average stream width will be determined from systematic width measurements taken in each zone. The number of intervals in each zone will depend on the length of the zone. Each measurement will be recorded at the appropriate location on the stream maps prepared in 1989 and 1990.

Weir camp crews will perform daily ground surveys of intertidal and upstream portions of the weired systems as well as 21 other pink and chum salmon spawning streams (Figure 2). Live and dead pink and chum salmon will be enumerated in standardized intertidal and upstream zones in each stream. During each stream survey the following data will be recorded:

1. anadromous stream number and name (if available);
2. latitude and longitude of the stream mouth;
3. date and time (24 hour military time);
4. tide stage;
5. observer names;
6. counts of live and dead salmon by species and tide zone (0.0-1.8m, 1.8-2.4 m, 2.4-3.0 m, and 3.0-3.7 m above mean low water and upstream) and;
7. weather and comments on visibility, lighting, and other survey conditions.

All data will be recorded on pre-printed data sheets. Maps will be improved and modified during the survey to show spawner distribution within each zone and the upstream limit of spawning.

Counts of live and dead salmon will be made for the five tide zones (the intertidal zones < 1.8 m, 1.8-2.4 m, 2.4-3.0 m, 3.0-3.7 m above mean low water and the upstream zone) from the 1.8 m tide level to the limit of upstream spawning on all 27 streams during daily surveys. Tide stage will be monitored continuously and survey times and direction will be adjusted accordingly. If the tide stage at the time of the walk is at or below the 1.8 m level the stream walk will begin at the stream mouth and progress upstream. The mouth or downstream

limit of the stream will be defined as the point where a clearly recognizable stream channel disappears or is submerged by salt water. Fish seen below the downstream limit will be included in an estimate of fish off the stream mouth and noted as a comment on the data form. If the intertidal portions of the stream above the 1.8 m level are submerged at the time the walk begins, the crew will go to the upstream limit of the walk, proceed downstream, and coincide the end of the walk with the time predicted for the tide to be at or below the 1.8 m level. The upstream limit of a walk will be determined by the presence of natural barriers to fish passage (i.e. waterfalls), by the end of the stream, or by the upstream limit of spawning. The upstream limit of spawning will be marked on U.S. Geological Survey color aerial photos of each stream following each survey.

For counts of live and dead fish on moderate size streams with a single channel, crew members will walk together but independently count live fish in each intertidal zone. Crew members will individually enter their count on mechanical hand tallies. A maximum of three replicate counts may be made in each zone at the request of either observer. Upstream counts in a single channel will be similarly conducted at convenient stopping points (i.e., log jams or other clear counting delineators). For large braided or branched streams, each crew member will count separate channels or upstream forks. To avoid confusion with counts of live fish, counts of dead fish will be recorded on the return leg of the stream walk. Only fish that have died since the previous count will be tallied as dead in the daily surveys. To prevent duplicate counts between surveys, tails and tags of all dead pink and chum salmon observed will be removed. To avoid perpetuating counting biases within a counting crew, personnel will be rotated between crews daily. When possible, crew members will not be assigned to the same streams on succeeding days.

Crews marking, measuring, and mapping tide zones will conduct foot surveys of the intertidal stream bed and adjacent beaches to document, map, and classify oil present. A composite sample of mussels will be collected at the mouth of each stream for hydrocarbon analyses. Results of the analyses will be used to document the level of oil impact that the stream sustained. Each sample will consist of enough mussels to provide 10 grams of tissue (approximately 30 mussels) for analysis. The mussels will be collected in the zone from 0-2 m above mean low water in the immediate vicinity of each stream mouth and will be collected above water to avoid contamination by hydrocarbons on the water surface. The samples from each stream will be stored in separate, properly cleaned, glass jars with teflon lined lids. Appropriate chain of custody forms will accompany each sample.

Stream-life Studies

All 27 streams in the ground survey program are included in a stream life study (Figure 2). Average stream residence (stream life) on these streams will be estimated using data from daily ground surveys already described. On 25 of these 27 streams a second, independent estimate of stream life will be made using tagging results similar to those described by McCurdy (1984) and Helle et al (1964). A third independent estimate of stream life will be made at the seven

weired systems using daily weir data and carcass counts from daily ground surveys.

In the tagging study, fish will be captured with beach seines at the stream mouths and tagged with Peterson disks. Tags will be uniquely colored to represent day of tagging and uniquely numbered for identification of individual fish and stream. Each week 120 fish will be tagged from each of 24 streams. At another streams, the largest stream in the study, 200 tags will be applied weekly. If fewer than the desired number of fish were available, all fish captured will be tagged. Tagged live and dead fish will be totalled by color within each tide zone during daily ground surveys and tags recovered from carcasses. Where possible individual tag numbers will be recorded for tagged live fish.

DATA ANALYSIS

Data analysis procedures are similar or identical to those used in NRDA F/S Study #1. These procedures have been scrutinized thoroughly by the NRDA peer review process and approved by the Management Team. Report formats will be in accordance with those established by the Management Team. Reporting style and conventions will otherwise be in accordance with the ADF&G Division of Commercial Fisheries style manual.

Aerial Survey Data

Annual, spawning escapement estimates (E) for pink salmon within each surveyed stream will be made using a geometric approach similar to that described by Johnson and Barrett (1986):

$$E = \frac{\sum \left[(J_i - J_{(i-1)}) L_i - \frac{(J_i - J_{(i-1)}) (L_i - L_{(i-1)})}{2} \right]}{S}$$

Where i = survey number,
 J_i = julian date,
 L_i = survey estimate of live fish in the stream on survey
 i ,
 S = stream life (in days).

If the maximum daily survey of live fish in the stream exceeds the total escapement estimate based on the geometric method, the maximum daily survey count is treated as the total escapement.

Total Escapement Enumeration Data

Total escapement for streams with weirs will be the summation of daily counts of fish through the weir. Live fish present in the stream on any date will be the difference between the cumulative count of live fish by that date and the cumulative carcass count by that date. Estimates of fish present were used to validate coincidental counts from aerial and ground surveys.

Ground Survey Data

Ground survey counts will be summarized by species, stream, survey date, the four intertidal zones and the upstream zone, and by observer for all 27 streams in the study. Spawning escapement to streams surveyed from the ground will be estimated using the geometric method described for aerial survey data. Frequently survey counts (L_1) will be replicated as paired observations from two observers walking in tandem. The escapement estimate for a section walked in tandem will be the mean of the observations. The variance will be estimated using all replicates for the section. A one way analysis of variance will be used to test for differences between replicate observations from separate observers. In instances where the maximum daily sum of live and dead fish in a stream exceeds the total escapement estimate for the stream based on the geometric method, the maximum daily sum of live and dead will be the total escapement estimate.

Stream-life Data

Tagging data will be used to calculate stream life values for individual fish as:

$$S = J_r - J_c$$

where J_c = julian date when the live tagged fish was first observed entering the stream channel from the milling area at the mouth.

J_r = julian date of tag recovery from the dead fish.

The stream life estimates for each stream and weekly strata will be the average for individual fish in the strata. The season-average stream life estimate will be the average of strata estimates. Stream life estimates within weekly time strata will also be averaged across all streams to examine time trends in stream life.

Another mean stream life estimate for each stream will be calculated as the difference between the mean date of abundance of new arrivals of live fish in the stream and the mean date of abundance of daily dead counts as follows:

$$S = \frac{\sum D_i J_i}{\sum D_i} - \frac{\sum [(L_i - L_{(i-1)}) + D_i] J_i}{\sum [(L_i - L_{(i-1)}) + D_i]}$$

where i = survey number,
 L_i = number of live fish observed on survey i ,
 D_i = number of dead fish observed on survey i ,
 J_i = Julian date of survey i .

For weired streams a third estimate of mean stream life based on daily counts of live fish through the weir and daily dead counts in the stream will be as follows:

$$S = \frac{\sum [(J_i - J_{(i-1)}) \sum (W_i - D_i)]}{\sum W_i}$$

Where i = serial day of weir operation,
 J_i = Julian date,
 W_i = live fish passed through the weir on day i ,
 D_i = count of dead fish in the stream on day i ,
 S = stream life (in days).

If observations for day i are missing, total live fish in the creek on day i ($\sum (W_i - D_i)$) will be linearly interpolated.

If significant differences occur in stream life estimates between streams or time strata, stream and week specific stream life estimates will be applied to similarly stratified aerial and ground observations when estimating escapements using the geometric method.

SCHEDULES AND PLANNING

Data Collection, Analysis and Reporting Schedule

Data collection, analyses and reporting of results for the 1991 field season will proceed as follows:

July 1 - 15 Sept. 1991 Weir installation and operation, ground surveys, and stream life studies. Inseason data entry of weir, ground survey, and aerial survey data. Analysis of inseason data and consultation with ADF&G Division of Commercial Fisheries management

personnel concerning management decisions regarding oil impacted stocks.

Sept 15 - 30 Nov. 1991 Completion of post season computer data entry and editing.

Sept 15 - 30 Dec. 1991 Completion of preliminary post-season data analysis and progress report.

Dec. 15 - 28 Feb. 1992 Finalize post season data analyses and project completion report.

Sample and Data Archival

All project operational plans, data logs, field notebooks, as well as original copies of draft and final reports will be kept in locked file storage in the ADF&G Commercial Fisheries Division and OSIAR offices in Cordova.

Weir data, ground survey, tagging, and tag-recovery forms will be labeled with a three part alpha-numeric code unique to each data type, stream and, date. At the end of each day, forms will be carefully edited and the code for each will be recorded in a data collection log maintained by each field crew. As forms are logged they will be initialed by the crew member doing the log in procedures for that day. Any biological samples collected will similarly be coded as to sample type, sampling site, and date. All data and samples collected will be remitted to the Cordova ADF&G office on a weekly schedule according to standard chain of custody procedures. Data collection log numbers, date sent and the initials of the person sending, will be recorded in a the field data camp data transmission log. Data received in Cordova will recorded in a data and sample transmission log which will show the codes assigned to each form and sample at each field camp as well as the date received and the initials of the receiver.

Original data forms for each data type and stream will be stored in separate, labeled three ring binders in the Oil Spill Impact, Assessment, and Recovery (OSIAR) office. Backup photocopies of the data will be stored in corresponding binders in the ADF&G Commercial Fisheries Division office in Cordova. All samples will be placed in locked storage and sent to the appropriate processing laboratories or centralized storage facilities when appropriate. Standard chain of custody procedures will be followed when any data or samples are remitted from the custody of project personnel in Cordova.

All data will be edited for errors immediately upon receipt in Cordova and then entered into a microcomputer data base in RBASE format. The RBASE data base will be accompanied by full documentation including a description of all columns, tables, and applications. Backup copies of the data base will be updated after every data edit or update and placed in locked, fireproof storage in the OSIAR and Commercial Fisheries Division offices. A complete log of data entries, edits, and archives will be maintained by project personnel which will reflect the alpha numeric data form codes, the date of entry or editing, and the initials of the person performing these functions.

Management Plan

The Principal Investigator (PI) for the project is a Fisheries Biologist III with the Alaska Department of Fish and Game. The PI will be responsible for writing project operational plans, administering project budgets, quality control of data collection, supervising data analyses and, co-authoring final reports. The PI will be assisted by a Fisheries Biologist II Project Leader (PL) who will hire project personnel, supervise day to day project operations, maintain data quality, assist in data analyses, and coauthor final reports. The PL will be assisted by two Fisheries Biologist I's. One of these assistants will be in charge of installing weirs and camps, weir operations and, remote camp logistics. The other assistant will supervise data collection activities in the ground survey and stream life studies. Each weir camp will be manned by four people one of whom will be funded by NRDA Study F/S #3 for recovery of adult salmon bearing coded-wire tags. The Each crew will have of one Fisheries Technician III crew leader. The remainder of each crew will be Fisheries Technician II's. Each day, two persons on each crew will tend the weir and conduct the ground survey, stream life, and tag recovery activities on the weired stream. The other two crew members will conduct ground survey, stream life and, tag recovery activities on the unweired streams.

Project Logistics

Weir and camp materials will be purchased in the Spring of 1991 with funds from NRDA F/S Study #1. The ADF&G R/V Montague will transport materials to the weir sites in June of 1991. Weirs and camps will be installed at seven sites (Figure) in the last week of June. Weir operations, ground surveys and, stream life studies will begin on July 1.

Weirs will be supplied semi-weekly by the R/V Montague or as needed by fixed wing aircraft. The PL and the assistant project leaders will visit each camp on a weekly schedule to oversee weir and camp operations, collect completed data forms and heads from tagged fish, answer questions from field crews, and monitor the data quality of data collected. The project leader or the assistant project leaders will maintain twice daily radio schedules with weir camps. During radio schedules, weir crew will transmit weir counts and stream walk counts to the Cordova office and transmit any other information or requests essential to camp operations. Data collected each week will be edited and entered into an RBASE data base in Cordova by a Fisheries Technician III. The PI and the PL in consultation with the OSIAR Biometrician will update escapement estimates based on aerial and ground survey data and weir counts. These analyses will be completed daily and the results will be passed on to the ADF&G Division of Commercial Fisheries PWS Area Management Biologist. In consultation with the PI, the PL and, other ADF&G fisheries management and research staff, the Area Management Biologist will use these results to make inseason fisheries management decisions.

BUDGET

Salaries	\$ 120.0
Travel	\$ 2.0
Contractual	\$ 43.0
Commodities	\$ 25.0
Equipment	\$ 40.0
Total	\$ 230.0

PERSONNEL QUALIFICATIONS

Fisheries Biologist III Principal Investigator - Samuel Sharr

Mr. Sharr 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.

Fisheries Biologist II Project Leader - Dan Sharp

Mr. Sharp has been employed by ADF&G since 1982. As a biologist for the ADF&G Susitna Hydroelectric Project Mr. Sharp gained valuable experience in a wide variety of techniques to enumerate salmon escapements and estimate migratory timing. His experience included operation of weirs, sonar counters and fishwheels and tagging studies for of juvenile and adult salmon. Mr. Sharp has been the Fisheries Biologist II Project Leader for the tagging portion of NRDA F/S Study #3 since its inception.

Fisheries Biologist I Assistant Project Leader - Stephanie Carpenter

Ms. Carpenter was a Fisheries Biologist I for NRDA F/S Study #1 in 1990 and supervised the installation and operation of four adult pink salmon weirs in PWS.

Fisheries Biologist I Assistant Project Leader - Mary Hausler

Ms. Hausler was a Fisheries Technician in the escapement ground survey portion of NRDA F/S Study #1 in 1989 and as a Fisheries Biologist I in 1990, she supervised the all ground survey and stream life studies for that project.

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