RPWG
JJ

VOLUME 2

DETAILED OPERATIONAL PLANS FOR STUDIES

IN THE

STATE/FEDERAL NATURAL RESOURCE DAMAGE ASSESSMENT PLAN FOR THE EXXON VALDEZ OIL SPILL

DRAFT

CONFIDENTIAL

Fisheries Studies 1 - 17

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	AW2	Injury to Subtidal
	AW3	Hydrocarbons in Water
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CONFIDENTIAL

STATE/FEDERAL NATURAL RESOURCE DAMAGE ASSESSMENT
DETAILED STUDY PLAN

DRAFT

Project Title:	INJURY TO SALMON SPAWNING AREAS IN PRINCE WILLIAM SOUND
Study ID Number:	Fish/Shellfish Study Number 1
Lead Agency:	State of Alaska, ADF&G; Commercial Fish Division
Cooperating Agency(ies):	Federal: USFS State: DNR
Principal Investigator:	Sam Sharr, Fishery Biologist
Assisting Personnel:	
Date Submitted:	October 11, 1989

	Signature	Date
Principal Investigator:	_____	_____
Supervisor:	C Meacham	10-11-89
Consulting Biometrician:	Brian L. Bue	Oct. 11, 1989
OSIAR Senior Biometrician:	_____	_____
OSIAR Program Manager:	_____	_____
OSIAR Director:	_____	_____

INTRODUCTION

The project is designed to evaluate possible changes in numbers and distribution of pink and chum salmon spawning in intertidal and upstream areas relative to oil contamination from the Exxon Valdez spill. A significant amount of pink and chum salmon spawning in Prince William Sound occurs in intertidal areas (up to 75% in some years). These intertidal areas are extremely susceptible to marine contaminants and may adversely affect spawner distribution and success in Prince William Sound.

The Alaska Department of Fish and Game performed spawning ground surveys of the major anadromous spawning streams in Prince William Sound from the late 1950's through 1985. Approximately 116 streams were surveyed at least once annually during the peak of spawning. Program funding was severely curtailed in 1987 and 1988; consequently, only 58 streams have been walked annually in recent years. This study will again expand the number of spawning streams surveyed in order to document the effect of the oil spill on pink and chum salmon intertidal spawning. The study will determine the post oil spill distribution of spawning, help in the evaluation of streams for inclusion in the Injury to Salmon Eggs and Pre-emergent Fry in Prince William Sound study (OSIAR study F/S 2) and the Salmon Coded-wire Tag Studies in Prince William Sound (OSIAR study F/S 3), and provide an atlas of aerial photographs and detailed maps for important spawning sites.

OBJECTIVES

1. Determine the presence or absence of oil on intertidal habitat used by spawning salmon.

2. Document the physical extent of oil distribution on intertidal spawning areas.

3. Estimate the numbers of spawning salmon by species within standardized intertidal and upstream zones for 140 streams in Prince William Sound.

4. Produce a catalogue of aerial photographs and detailed maps of spawner distribution for each stream sampled.

5. Identify potential alternative methods and strategies for restoration of lost use, populations, or habitat where injury is identified.

METHODS/DATA ANALYSIS

Study Design

Three crews of two people each will perform foot surveys of intertidal and upstream portions for 140 major pink and chum salmon spawning streams. Each stream will be surveyed three times at approximately two week intervals during the spawning season.

Streams to be surveyed will be selected according to the following criteria:

1. The stream is included in the aerial survey program.
2. All streams in the pink and chum salmon egg deposition and pre-emergent fry projects will be included.
3. Streams enumerated in prior spawning ground foot survey programs will be included.
4. Streams utilized by salmon stocks from the early, middle and late run stocks will be included.

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 (0.0-1.8 m, 1.8-2.4 m, 2.4-3.0 m, and 3.0-3.7 m above mean low water and upstream);
7. Weather and comments on visibility, lighting, and other survey conditions.

All data will be recorded on pre-printed mylar data sheets which will overlay a map of the stream. Stream maps will be drawn from three sets of aerial photographs prior to the survey. Maps will be improved and modified during the survey to show the location of tide zones, stakes and key land marks for identifying zones, spawner distribution within each zone, and the upstream limit of spawning. Particular attention will be given to spawner density and distribution observations for the 46 streams to be sampled during the Pink and Chum Salmon Pre-emergent Fry Sampling project (OSIAR study F/S 2).

Streams will be surveyed in a systematic order around Prince William Sound. The ADF&G R/V Montague will house and support three survey crews of two people each. Each two man crew will use a skiff for transport between the R/V Montague and the survey streams.

On the first circuit of Prince William Sound one crew of two people will measure and mark tide levels at each stream to be enumerated. The stream bed location of tide levels 1.8, 2.4, 3.0, and 3.7 meters above mean low water will be determined using a professional grade surveyors level and will be marked with colored coded surveyors stakes (orange, blue, green, and white, respectively). The bench mark for measuring the tide levels at each stream will be based on the tide level at the time of the survey. Location and time specific tide levels

will be obtained from a commercial tide stage computer software package run on a microcomputer aboard the R/V Montague. The tide level will be relayed to the shore based crew via radio.

During the first survey circuit, a composite sample of mussels (Mytilus sp.) will be collected at the mouth of each stream for hydrocarbon analysis. Results of the analysis will be used to document the level of oil impact sustained by the stream. Each sample will consist of enough mussels to provide 10 grams of tissue (approximately 30 mussels) for analysis. The mussels will be collected 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. Samples from each stream will be stored in separate glass jars with teflon lined lids. Each jar and lid will be pre-rinsed three times with dicloromethane, dried, and kept in locked storage prior to use. Care will be taken not to contaminate the jar or lid prior to or during sampling. Each sample jar will be neatly labeled with indelible ink or pencil on "Rite-In-The-Rain" paper. The label will bear a sample number, sampling date and time, tide stage, species, the ADF&G stream name and number, the stream mouth latitude and longitude, and the sampler name(s). The samples will be stored in the freezer aboard the R/V Montague until the boat reaches port. At that time they will be moved to locked shore based freezer storage. Appropriate chain of custody forms will accompany each sample.

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 140 streams for all three circuits. 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 mouth of the stream 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 when the walk begins, the crew will proceed to the upstream limit of the walk, walk downstream, and coincide the end of the walk with the time predicted for the tide to 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 (ie. 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.

Count of live and dead fish on each stream will be done by a crew of two technicians or biologists. For streams of moderate size and having a single channel the crew members will walk together but independently count live fish. Each member will tally their count on a mechanical tallywhacker. At the end of each zone the two crew members will compare counts. If both are comfortable with their counts and the difference between the two is less than 10% of the lower of the two counts for the zone, the two counts will be averaged. If the two counts differ by more than 10% of the lower of the two counts both surveyors will re-count the zone until their counts differ by 10% or less. Upstream counts in a single channel will be similarly compared at convenient stopping points (ie. log jams or other clear counting delineators). To avoid confusion with counts

PROJECT BUDGET [1]

Line Item	Category	Budget
100	Personnel Services	$ 52,200
200	Travel	$ 5,500
300	Contractual	$ 58,100
400	Commodities	$ 17,900
500	Equipment	$ 11,100
700	Grants	$ 0
Total		$144,800

[1] Budget is for all activities performed from March 27, 1989 to February 28, 1990.

FUNDED PERSONNEL

Class	PCN	Name	PFT_mm	SFT_mm
FB II	11-		0	
FB I	11-			6
FT III	11-			2
FT II	11-			4
FT I	11-			2

LITERATURE CITED

Helle, J.H., R.S. Williamson, and J.E. Bailey. 1961. Intertidal ecology and life history of pink salmon at Olsen Creek, Prince William Sound, Alaska. U.S. Fish and Wildlife Service S.S.R. -Fisheries No. 483.

Johnson, B.A., and B. Barrett. 1988. Estimation of salmon escapement based on stream survey data. Alaska Department of Fish and Game, Division of Commercial Fisheries, Regional Information Report No. 4K88-35, Kodiak.

McCurdy, M.L. 1984. Eshamy district pink salmon streamlife study, 1984. Alaska Department of Fish and Game, Division of Commercial Fisheries, Prince William Sound Area Data Report, 84-18, Cordova.

CONFIDENTIAL

STATE/FEDERAL NATURAL RESOURCE DAMAGE ASSESSMENT
DETAILED STUDY PLAN

DRAFT

Project Title:	INJURY TO SALMON EGGS AND PRE-EMERGENT FRY IN PRINCE WILLIAM SOUND
Study ID Number:	Fish/Shellfish Study Number 2
Lead Agency:	State of Alaska, ADF&G; Commercial Fish Division
Cooperating Agency(ies):	Federal: USFS State: DNR
Principal Investigator:	Sam Sharr, Fishery Biologist
Assisting Personnel:	
Date Submitted:	October 11, 1989

	Signature	Date
Principal Investigator:		
Supervisor:		10-11-89
Consulting Biometrician:		Oct. 11, 1989
OSIAR Senior Biometrician:		
OSIAR Program Manager:		
OSIAR Director:		

INTRODUCTION

The project is designed to evaluate possible changes in pink and chum salmon egg to pre-emergent fry survival in intertidal and upstream areas relative to oil contamination from the Exxon Valdez spill. A significant amount of pink and chum salmon spawning in Prince William Sound occurs in intertidal areas (up to 75% in some years). These intertidal areas are extremely susceptible to marine contaminants and may adversely affect spawner distribution and success in Prince William Sound.

The Alaska Department of Fish and Game has performed egg and pre-emergent fry digs at some of the approximately 900 anadromous fish streams in Prince William Sound since the early 1960's (Pirtle and McCurdy 1980, McCurdy 1984). Pre-emergent fry digs provide an abundance index for pink salmon which is used to forecast future pink salmon returns. As many as 45 streams were sampled annually prior to 1985. Since then only 25 index systems considered representative of pink and chum salmon producing streams in Prince William Sound have been sampled. The oil spill has the potential to cause mortality to the critical egg and fry life stages and thus an increased and more comprehensive egg/fry dig program is called for. This project is designed to meet this need by assessing the effect of the oil spill on egg to fry development of wild pink and chum salmon stocks.

OBJECTIVES

1. Estimate mortality of pre-emergent fry in oiled and non-oiled streams immediately following the Exxon Valdez oil spill of March 24, 1989.

2. Estimate over winter survival (eggs to pre-emergent fry) of pink and chum salmon eggs in oiled and non-oiled areas.

3. Assess loss in production, if any, from changes in overwinter survival.

4. Use tissue samples from pre-emergent fry and from mussels (Mytilus sp.) to document hydrocarbon contamination in streams where oil is not evident visually.

5. Identify potential alternative methods and strategies for restoration of lost use, populations, or habitat where injury is identified.

METHODS/DATA ANALYSIS

Sample Sites

Fry sampling was performed on two occasions immediately after the oil spill in an effort to assess initial fry abundance and possible fry mortality just prior to and immediately after oil impact. The second sampling trip was also used to evaluate streams in the areas impacted by oil for egg/fry dig potential. Thirty nine streams throughout Prince William Sound were sampled during the first trip. The second trip occurred approximately two weeks after the oil spill and concentrated on the Central and Southwest areas of Prince William Sound. During the second event 14 streams were resampled (representing both oiled and non-oiled areas) and an additional 17 streams were surveyed to assess their potential as egg and pre-emergent study streams (Figure 1, Appendix A).

Spring fry digs will be conducted at 56 streams. The 25 streams presently studied will be sampled to allow for a comparison of abundance data after the oil spill with historic data. Of these streams, 20 received no oil impact, 3 are suspected of having received some impact, and 2 have visual evidence of oil impact at the stream mouth. An additional 21 streams will also be sampled. These streams were selected from areas with no oil impact to high impact in Central and Southwest Prince William Sound where the majority of the oiling occurred. New streams were selected using the following criteria:

1. Sufficiently large adult salmon returns to indicate a high probability of success in egg/fry digging.
2. Past history of egg/fry digging.
3. Streams which had low to no oil impact in the immediate vicinity of high oil impact streams. This will help account for possible variability due to differing climatic/stream conditions.

Based on these criteria, the added sample streams have the following characteristics: two have a history of fry sampling and are in areas with high probability of oiling, 11 will have no history of fry sampling and are in areas with high probability of oiling, two will have a history of fry sampling and are in areas with little oil impact, and six will have no fry sampling history and are in areas with little oil impact (Appendix A).

Fall egg digs will be conducted on a subset of 31 of the 56 streams sampled for pre-emergent fry. Egg dig streams have the following characteristics: eight are in areas with heavy oil impact, five are in areas of moderate oil impact, four are in areas where oil impact is suspected but the extent of contamination is unknown, and 14 will be in areas suspected of receiving little or no oil.

Sample Design

Sampling will be conducted in two phases: egg-digs in October, and pre-emergent fry digs in mid-March to mid-April. Sampling methods are identical for the egg and pre-emergent fry digs. On each sample stream, four zones, 3 intertidal and one above tidal inundation, will be identified and marked by crews conducting stream surveys under Fish/Shellfish Study Number 1 (Injury to Salmon Spawning Areas in Prince William Sound). The zones are 1.8-2.4 m, 2.4-3.0 m, 3.0-3.7 m

Pre-emergent fry/egg data will be summarized by date, stream, level of hydrocarbon impact, stream zone, and number of live and dead, fry and eggs. A mixed effects analysis of covariance will be used to test for differences in 1) egg to fry survival due to oiling using the 31 streams sampled for both eggs and pre-emergent fry and 2) fry mortality immediately after oiling using the 14 streams which were replicate sampled in April 1989. Degree of oiling and height in the tidal zone will be treated as fixed effects. Height in the tidal zone will be nested within stream, a random effect. Covariates will be provided by hydrocarbon analysis of mussel populations in close proximity to each stream (collected by OSIAR study F/S 1).

If no suitable hydrocarbon data are available, analysis of variance will be used. Degree of oiling as visually assessed by the mapping portion of the Injury to Salmon Spawning Areas in Prince William Sound study (OSIAR study F/S 1) will be used to post-stratify streams. Degree of oiling and height in the tidal zone will again be treated as fixed effects. Height in the tidal zone will be nested within stream, a random effect.

An assessment of lost fry production will be made if differences in egg to fry survival due to oiling are detected. Average survival from unoiled areas will be used to estimate potential fry density in oiled areas. Observed and potential fry densities will then be expanded to estimate total observed and potential fry. The difference between the two estimates will be considered lost fry production.

Specific statistics to be estimated are:

1. Number of dead and viable eggs per square meter by salmon species, stream, and stream zone.
2. Number of dead and live fry per square meter by salmon species, stream, and stream zone.
3. Egg to fry survival by salmon species, stream, and stream zone.
4. Lost production by salmon species, stream, and stream zone.

SCHEDULES AND REPORTS

Date(s)	Activity
Mar. 27 - Ap. 7 1989	Fry digs in 31 sample streams (first pass).
Ap. 14 - Ap. 22 1989	Fry digs and stream surveys in 31 streams (second pass).
November 1 - 15	Egg digs in 31 test streams.
Apr. 30 - Dec. 15 1989	Laboratory work, fry and egg counts.
June - December 1989	Data entry and analysis
January 1989	Preliminary report on impacts of oil egg and fry survival.
March 1 1990	Prepare final study plan for the 1990 field season.
March 15 - Ap. 15 1990	Fry digs on 46 study streams.

PROJECT BUDGET[1]

Line Item	Category	Budget
100	Personnel Services	$ 82,200
200	Travel	$ 5,000
300	Contractual	$ 16,200
400	Commodities	$ 5,700
500	Equipment	$ 40,000
700	Grants	$ 0
Total		$149,100

[1] Budget is for all activities performed from March 27, 1989 to February 28, 1990.

FUNDED PERSONNEL

Class	PCN	Name	PFT_mm	SFT_mm
FB II	11-		12	
FT III	11-			4
FT III	11-			4
FT II	11-			4
FT II	11-			4
FT I	11-			4
FT I	11-			4

LITERATURE CITED

McCurdy, M.L. 1984. Prince William Sound General Districts 1976 pink (Oncorhynchus gorbuscha) and chum (O. keta) salmon aerial and ground escapement surveys and consequent brood year egg deposition and pre-emergent alevin surveys, brood years 1980 through 1983. Alaska Department of Fish and Game, Division of Commercial Fisheries, Technical Data Report No. 116, Juneau.

Pirtle, R.B. and M.L. McCurdy. 1980. Prince William Sound General Districts 1976 pink (Oncorhynchus gorbuscha) and chum salmon (O. keta) aerial and ground escapement surveys and consequent brood year egg deposition and pre-emergent fry index programs. Alaska Department of Fish and Game, Division of Commercial Fisheries, Technical Data Report No. 51, Juneau.

Whittier

PRINCE WILLIAM SOUND

FRY DIG SITES - 1989

Figure 1. Location of streams and sampling sites in Prince William Sound, 1989.

Appendix A. Pink and Chum salmon egg and pre-emergent fry dig locations.

Stream Number	Stream Name	Stream Location	Current Fry Index	1989 dig 1st Pass	1989 dig 2nd Pass	Proposed Digs	
						Fry	Egg/1
11	Humpback Creek	Orca Inlet	X	X		X	
35	Knoppen Creek	Sheep Bay	X	X		X	
51	Control Creek	Port Gravina	X	X		X	X-c
80	Whalen Creek	Port Fidalgo	X	X		X	X-c
87	Sunny River	Port Fidalgo	X	X		X	
104	None	Tatitlek Narrows/Virgin Bay		X			
106	Gladough Creek	Tatitlek Narrows/Virgin Bay		X			
107	Black Creek	Tatitlek Narrows/Virgin Bay		X			
117	Indian Creek	Galena Bay	X	X		X	
121	Gregorieff Creek	Jack Bay	X	X		X	
153	Stellar Creek	Sawmill Bay	X	X		X	
262	Cutthroat Creek	Unakwik Bay	X	X		X	
276	Blackbear Creek	Eaglek Bay	X	X		X	X-c
322	Coghill River	Port Wells	X	X		X	
421	Mill Creek	Bettes Bay	X	X		X	X-c
430	Meacham Creek	Pigot Bay	X	X		X	
455	Paulson Creek	Cochrane Bay	X	X	X	X	X-c
480	Mink Creek	Mink Harbor/Port Nellie Juan	X	X	X	X	X-c
485	None	West Finger Inlet/Port Nellie Juan			X	X	X-c
495	Chimevisky Creek	McClure Bay/Port Nellie Juan			X	X	X-c
506	Loomis Creek	North of Eshamy Bay			X	X	X-o
604	Erb Creek	Ewan Bay/Dangerous Passage	X	X	X	X	X-o
618	None	Chenega Island - East Shore			X	X	X-o
621	Totemoff Creek	Chenega Island - West Shore		X	X	X	X-o
628	None	Chenega Island - East Shore			X	X	X-o
630	Bainbridge Creek	Whale Bay - West Arm			X	X	X-c
632	Claw Creek	Whale Bay	X	X	X	X	X-o
637	None	Point Countess outside of Whale Bay			X	X	X-o
653	Hogg Creek	Hogg Bay/Bainbridge Island			X	X	X-c
663	None	Shelter Bay/Evans Island			X	X	X-o
665	None	Evans Island				X	X-o
666	O'Brien Creek	Evans Island				X	X-c
673	Falls Creek	Latouche Island/West Shore	X	X	X	X	X-o
677	Hayden Creek	Latouche Island/West Shore		X	X	X	X-o
678	None	Sleepy Bay/North end Latouche Island			X	X	X-o
681	None	Hogan Bay/Knight Island		X	X	X	X-o
680-2	None	Mummy Bay/Northeast Arm, Knight Island			X		
682	None	Snug Harbor/Knight Island		X	X	X	X-o
684-1	None	Marsha Bay/Knight Island		X			
687	None	Bay of Isles/Knight Island		X			
689	None	Louis Bay/Knight Island		X			
692	None	Herring Bay/Knight Island		X	X	X	X-o
695	None	Drier Bay/Knight Island			X	X	X-c
699	None	Drier Bay/Knight Island			X	X	X-c
707	McCleod Creek	McCleod Harbor/Montague Island		X			
711	Hanning Creek	Hanning Bay/Montague Island		X			
740	Kelez Creek	Montague Island/Northwest Shore			X	X	X-c
744	Wilby Creek	Port Chalmers/Montague Island	X	X		X	
747	Cabin Creek	Port Chalmers/Montague Island		X			
749	Shad Creek	Port Chalmers/Montague Island	X	X	X	X	
759	Rocky Creek	Rocky Bay/Montague Island			X		
775	Pautzke Creek	Zaikof Bay/Montague Island	X	X		X	
788	Green Creek	Green Island			X	X	X-o
814	Constantine River	Port Eches/Hinchinbrook Island	X	X	X	X	
828	Cook Creek	Anderson Bay/Hinchinbrook Island	X	X	X	X	
850	Canoe Creek	Canoe Pass/Hawkins Island	X	X	X	X	
861	Bernard Creek	Windy Bay/Hawkins Island	X	X	X	X	X-c
Total Number of Streams			25	39	31	46	31

/1 o = Oiled; c = Control (not oiled)

Alaska Department of Fish and Game

Pre-emergent Pink and Chum Salmon Data Form

PAGE____ OF ____ FOR STREAM

_____

Field 1				2	3	4	5	6	7	8	9
DATE				DIST. NO.	SUB-DIST.	STREAM NO.	STUDY AREA	STRM AREA	TEMP.	CREW LEADER	DIG TYPE
MO.	DAY	YR.									

10	11	12	13	14	15	16	17	18	19	20	21	22	23	24
LOCATION	SAMPLE POINT	LIVE PINK EGGS	LIVE PINK FRY	PERCENT ABSORB.	DEAD PINK EGGS	DEAD PINK FRY	LIVE CHUM EGGS	LIVE CHUM FRY	PERCENT ABSORB.	DEAD CHUM EGGS	DEAD CHUM FRY	REMARKS	CONDITION	

11-CF-11

Appendix B. Field date form for pink and chum salmon egg and pre-emergent fry dig.

Pre-emergent Fry and Egg Deposition Pink and Chum Salmon

Data Form Codes

Field No. 10 - Location Code 1/

NIT 4' - 6'	=	Ø1Ø
NIT 6' - 8'	=	Ø2Ø
NIT 8' - 10'	=	Ø3Ø
NIT 10' - 12'	=	Ø4Ø
NIT 12' - 15'	=	Ø5Ø
NUPST	=	Ø6Ø
OIT 4' - 6'	=	Ø7Ø
OIT 6' - 8'	=	Ø8Ø
OIT 8' - 10'	=	Ø9Ø
OIT 10' - 12'	=	1ØØ
OIT 12' - 15'	=	11Ø
OUPST	=	12Ø

Field No. 10 - Sublocation code for far right digit in the above location Codes.

Ø = Where only one channel exists in sample location.

1 = Right hand channel facing upstream.

2 = Left hand channel facing upstream.

3 = Second (2nd) sample zone in same location.

4 = Third (3rd) sample zone in same location.

5 = Fourth (4th) sample zone in same location.

1/ If a sample location is known as OIT 9' the code would be the same as OIT 8' - 10'. This applies to all such sample location labels.

Appendix B.

Codes for Field 10 on the pink and chum salmon egg and pre-emergent fry data form.

INSTRUCTIONS

PINK AND CHUM SALMON DATA FORM

General Instructions

1) When the same number is to be used throughout a column or portion of a column, draw a vertical arrow.

2) Leading zeros and zero entries need not be entered, but numbers should be right centered.

Specific Instructions

Field 5 Study Area (Numerical order for area sampled within a stream).
Field 6 Stream Area (Type of area sampled - 1 for UPSTREAM with no intertidal, 2 for DOWNSTREAM with intertidal, 3 for INTERTIDAL, 4 for OLD UPSTREAM, 5 for NEW UPSTREAM, 6 for OLD INTERTIDAL, and 7 for NEW INTERTIDAL).
Field 7 Temperature (With decimal for Centigrade and without decimal for Fahrenheit).
Field 8 Crew Leader (0 for UNKNOWN, 1-199 for Southeastern, 200-299 for Prince William Sound, 300-399 for Cook Inlet, 400-499 for Kodiak, 500-599 for Chignik and Alaska Peninsula, 600-699 for A-Y-K Region).
Field 9 Dig type (0 or 1 for spring pre-emergent and 2 for fall egg deposition).
Field 10 Location (Sample subgroup of same riffle or group of similar riffles).
Field 11 Sample Point (Non-identical numbered samples within a location).
Field 14 Percent Absorbed (Average percent frysac absorbed for pink salmon).
Field 19 Percent Absorbed (Average percent frysac absorbed sor chum salmon).
Field 22 Remarks (1 for Sculpin, 2 for Flatworms, 3 for Roundworms, 4 for Dolly Varden, 5 for Copepods, and 6 for Flounders).
Field 23 Condition (1 for NOT SAMPLED DUE TO SNOW, 2 for NOT SAMPLED BECAUSE DRY, and 3 for NOT SAMPLED DUE TO WEATHER).

Appendix B.

Codes and explanations for fields 5-11, 14, 19, 22, and 23 on the pink and chum salmon egg and pre-emergent fry data form.

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STATE AND FEDERAL NATURAL RESOURCE DAMAGE ASSESSMENT
DETAILED STUDY PLAN

Project Title:	Code-wire Tag Studies on Prince William Sound Salmon
Study ID Number:	Fish/Shellfish Study Number 3
Lead Agency:	State of Alaska, ADF&G.
Cooperating Agencies:	NOAA, UA
Principle Investigators:	Sam Sharr, Commercial Fish Div. Larry Peltz, FRED Division
Assisting Personnel:	Hal Geiger, Karen Crandall
Date Submitted:	October 13, 1989

DRAFT

	Signature	Date
Principal Investigator:	_____	_____
Supervisor:	_____	_____
Consulting Biometrician:	_____	_____
OSIAR Biometrician:	_____	_____
OSIAR Program Manager:	_____	_____
OSIAR Director:	_____	_____

INTRODUCTION

The oil spill in Prince William Sound may cause differential survival of various salmon stocks in Prince William Sound by several mechanisms. The Alaska Department of Fish and Game (ADF&G), cooperating with the National Marine Fisheries Service (NMFS), and the University of Alaska (UA), has proposed a series of studies to measure the effects of the oil spill on salmon. A part of this series of studies is an aggressive coded-wire tagging effort on every species of salmon in Prince William Sound. The tagging will involve both pink and sockeye salmon from wild stocks, and coho, chinook, pink, and sockeye salmon from hatcheries, although the study will intensively focus on pink salmon.

Estimates of the loss of salmon production will come from three sources. The first source will be direct comparisons among coded-wire tagged stocks with different exposures to the toxicants. The second source will be the deviation of post-spill production (catch and escapement) from estimates of historical production. Historical production estimates will be based on historical aerial surveys of spawners adjusted with results from stream life studies conducted in concert with this coded-wire tagging project. Finally, the most powerful estimate of loss of production will be by directly observing the exposure at each particular life stage for a particular stock group, then applying a priori survival estimates that can be tied into a particular dose-response relationship. As an example, suppose rearing juveniles are observed in habitat with known concentrations of toxic oil residuals. By knowing the stock origins of these juveniles, through tagging, a decrease in survival can be inferred from the results of laboratory studies. This decrease may not be detectable by observing survival in wild stocks because of natural variability.

Separate early marine life history studies conducted by ADF&G, UA, and NMFS will document the exposure history of each coded-wire tag lot. Stream life studies, conducted as a part of the coded-wire tagging of wild pink salmon stocks, will be used with information from aerial surveys of spawning areas and harvest records to estimate the total (post-oil spill) production of pink salmon. Studies of intertidal spawning areas and fry survival, conducted independently of the coded-wire tagging studies, will track fry production of pink salmon in each year of the project from both oiled and unoiled spawning areas. By applying estimates of marine survival developed from the tag studies to estimates of fry production from other studies, estimates of salmon production will be developed independently of the aerial survey and stream life studies.

OBJECTIVES

A. To estimate the marine survival rates and harvests of wild pink salmon from three streams with oil-contaminated estuaries and two streams with uncontaminated estuaries.

B. To estimate the marine survival rates and harvests of sockeye salmon from two watersheds with contaminated estuaries, and one with an uncontaminated estuary.

C. To estimate the marine survival rates and harvests of pink, chum, coho, sockeye, and chinook salmon released from the five hatcheries in the Sound; two of these hatcheries are in heavily oiled areas, while three are not.

D. To estimate the abundance of sockeye salmon smolts emigrating from the study streams.

E. To estimate the extent of straying of returning salmon in outlying areas.

F. Identify potential alternative methods and strategies for restoration of lost use, populations, or habitat where injury is identified.[1]

In addition to these objectives a major task of the project will be to provide marked salmon of known stock origin and exposure history to the ecosystem for the benefit of researchers sampling rearing juvenile salmon (Fish Study No. 4), and returning adult salmon in Prince William Sound.

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[1] Methods to accomplish this objective are not addressed in the plan. They will be developed only if injury is documented.

METHODS

Hatchery Pink and Chum Salmon

Fry from each of the four private non-profit (PNP) hatcheries in Prince William Sound will be captured at various times throughout emergence at approximately the same rate they are released from the incubators. They will be anesthetized in MS-222, their adipose fin excised, and a tag applied using Northwest Marine Technology equipment and tags. Following tagging, overnight tagging mortality will be assessed and a sample of fish will be dissected to document tag placement. The tagged fry will then be transported to small holding areas. They will be observed and fed for 72 hours after the last fish are added to the holding areas. After the holding period, the tagged fish will be mixed with their untagged cohorts to rear for the remainder of their hatchery residency. Total fry abundance in the population will be assessed using electronic counters at the time the fry are ponded. Finally, a written description of the tagging will be developed by Larry Peltz of ADF&G. This 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 coordinates coded-wire tagging in Alaska.

The four hatcheries will release thirteen groups of pink salmon in 1989. Only one of these groups will not contain tagged fish (Main Bay Hatchery pink salmon from AFK stock, produced at the Cannery Creek Hatchery). Each of the hatchery pink salmon tag groups will contain tagged fish at the rate of approximately 1.7 ppt (or about 1 tagged fish in each 570 hatchery fish released). The tag rate will be held constant across release groups to prevent confusion of differential tag mortality with variation in survival between release groups (see ADF&G Technical Fishery Report by Peltz and Geiger, In Press; or Geiger and Sharr, In Press). In 1989 chum salmon will be tagged at the rate of 17 ppt at the Solomon Gulch Hatchery near Valdez.

In 1990 hatchery pink and chum salmon tagging will continue at the same level of effort with the addition of chum salmon at the Esther Island Hatchery; approximately 250,000 of these chum salmon will be tagged in one release group using the methods outlined above for hatchery pink salmon. Tagging at the 1990 level for both pink and chum salmon will continue until 1993.

Wild Stock Pink Salmon

In 1990 wild pink salmon will be tagged from five stocks chosen to fit in with the a fry and egg dig survey (Fish Study No.2). The wild pink salmon tagging is experimental, and will not be a routine application of coded-wire tag technology. Fry will be captured as they emerge using various means. The fry will be anesthetized with MS-222 and tagged with Northwest Marine Technology tagging equipment and tags. The anesthesia and associated trauma will 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 attributed to other groups of salmon will be assessed at the time of recovery. The 1990 level of tagging is expected to continue until 1993.

Hatchery Chinook and Coho Salmon

Prior to tagging, smolt in hatcheries will be crowded using seines. A sample of smolt will be drawn from each rearing appliance in approximate proportion to the number of fish in that appliance. They will be anesthetized with MS-222, their adipose fin excised, and a tag applied using Northwest Marine Technology equipment and tags. A sample of fish from each day's tag production will be retained to estimate short-term tag loss and tag induced mortality. Following tagging, the tagged fish will be returned to mix with untagged cohorts. All mortalities during the first week after tagging will be examined, and the tag status will be noted. At the end of a week, the fish will again be crowded, and a sample of approximately 2,000-4,000 fish from each rearing appliance will be drawn. These fish will be anesthetized, and run through a tag detector. Petersen abundance estimates for all rearing appliances will be performed, and any major discrepancies from hatchery inventory records noted. Finally, a written description of the tagging will be developed by Larry Peltz of ADF&G. This 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 coordinates tagging in Alaska. Tagging of chinook salmon will begin in 1990 and is expected to be continued until 1993. The tagging of coho salmon began in 1989 and will be continued until 1993.

Sockeye Salmon

In 1989 wild sockeye salmon will be tagged from at Eshamy and Coghill Lakes. Smolt will be captured in traps as they migrate to saltwater. The smolts will be anesthetized with MS-222 and tagged with Northwest Marine Technology tagging equipment and tags. The anesthesia and associated trauma will require that the tagged fish

be held separate from their untagged cohorts until they appear 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 attributed to other groups of salmon will be assessed at the time of recovery. The rate of tag occurrence in the stock will be determined from counts at an adult salmon weir in each of the systems. Heads from fish with adipose fin marks will be taken at the weir and the tags decoded. Hatchery produced sockeye salmon smolts will be tagged using the methods described for chinook salmon above. The hatchery tagging will begin in 1989 and be continued until 1993. In 1990 a weir will be placed on the Jackpot system; tagging will begin here in 1990 and also be continued until 1993.

Adult Recovery Survey

From 1989 through 1999 the Alaska Department of Fish and Game will oversee the recovery of coded-wire tagged fish in commercial harvests of salmon in Prince William Sound. The recovery samples are from a stratified sample (Cochran 1977). Fisheries will be stratified by district and within each district into discrete time segments. The recovery will be further stratified by processor strata as described in Peltz and Geiger (In Press). For each time and area specific stratum, 20% of the catch 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, three facilities in Valdez, and one facility in Kodiak. When feasible, sampling will occur at additional facilities in Kodiak, Kenai, Anchorage, and Whittier. If feasible large floating processors will be sampled as needed. All deliveries by fish tenders to these facilities will be monitored by radio and by daily contact with processing plant dispatchers to insure 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 broodstock tag recovery effort at each of the three hatchery facilities where tags were initially applied. A minimum of 50% of the daily broodstock requirements of each facility will be scanned for fish with missing adipose fins. Finally, there will be an intensive survey of the adult pink salmon returning to natural systems where tagging was conducted, and a weir will be operated for sampling adult sockeye salmon on those systems where sockeye salmon were tagged.

In the catch, terminal harvest, broodstock, and natural system surveys, the total number of fish scanned and the total number of fish with missing adipose 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.

Tag Processing Laboratory

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 database. 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. Sampling information and tag codes entered into the database will be available for analysis the following morning. Data will be automatically transferred from Juneau to Cordova. Eventually, on-line access from Cordova will provide in-season information to fisheries managers in Cordova to allow assessment of oil spill impacts and implementation of any required in-season management actions. Catch and sampling information will be integrated with tag codes to automatically calculate in-season and post-season hatchery contribution estimates. A historic database of coded-wire tag information from Prince William Sound tagging and tag recovery programs will be maintained and will be easily accessible by managers and researchers.

Data Analysis

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 broodstock. For the hatchery stocks, 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 Prince William Sound: Peltz and Geiger (In Press), and Geiger and Sharr (In Press). Additional references on methods of tagging pink salmon in Prince William Sound can be found in Peltz and Miller (1988). In the case of the wild stocks, the methods and estimators and necessary assumption are described by Geiger (1988).

The basic principle behind the estimates can be described as follows. The contribution of a particular tag lot, to a particular fishery stratum, is estimated by 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 (broodstock) of each tag lot will be 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 (broodstock), and dividing by the estimated number of fish represented by the tag code.

This process can be expressed more formally as follows. The proportion of the fish tagged in tag group t (t=1,2,...,T) is denoted as P^t. Let N_i denote the number of fish caught in fishery strata i (i=1,2,...,k), let s_i denote the number of fish in that the fishery sampled for marks, and let x^t_i denote the number of tags recovered with code t in fishery i. The number of fish from the tag group t that were caught in the commercial fishery, C^t, is estimated as follows:

$$C^t = \sum_i x^t_i \, (N_i/s_i) \, P^{t-1}$$

The assumptions necessary to estimate C and the associated confidence intervals are as follows:

(1) the numbers of tagged and untagged fish are known exactly;

(2) the tagged sample of the original hatchery tag group is a simple random sample;

(3) the tags do not affect the fish with respect to the items under study (survival, timing, homing, etc.);

(4) none of the marks are lost;

(5) the number of fish in the fishery and the number of fish in the fishery sample are known exactly;

(6) the sample of the fishery is a simple random sample (i.e. every fish in the collection of fish under consideration has an exactly equal probability of selection independent of every other fish in the sample); and

(7) all marks are observed and all tags decoded.

Reporting

After the raw data has been posted to the central coded-wire tag data base in Juneau the Cordova area staff will create in-season data summaries and a post-season final summary. The results of these analyses will be completed and published in ADF&G Technical Data Reports, as well as in other technical and professional literature after oil spill litigation is completed. A draft report following guidelines set by the Trustee Council will be completed by December with the final report completed by the January following the recovery.

SCHEDULE

DATES	ACTIVITY
March 1 to May 1	Hatchery Tagging
March 20 to May 1	Wild Pink Tagging
May 1 to June 15	Wild Sockeye
October 1	Tag Application Report
June 20 to September 20	Catch Sampling
December 21	Overall Report

BUDGET/PERSONNEL

PROJECT BUDGET[1]

LINE ITEM	CATEGORY		BUDGET
100	Personnel Services	$	898.4
200	Travel	$	20.7
300	Contractual	$	528.8
400	Commodities	$	87.9
500	Equipment	$	407.1
700	Grants	$	0.0
TOTAL		$	1943.4

[1] Budget is for all activities performed from March 27, 1989 to February 28, 1990.

Appendix A

1990 Coded Wire Tagging Goals for Prince William Sound

Hatchery tagging

Hatchery	Species	Projected Release	Valid Tag Goal	Total Release /Marked Ratio
Armin F. Koernig	Pink	120,000,000	200,000	600
Cannery Creek	Pink	135,000,000	225,000	600
Solomon Gulch	Pink	120,000,000	200,000	600
Wally Norenburg	Pink	250,000,000	416,667	600
GRAND TOTAL	Pink	625,000,000	1,041,667	600
Solomon Gulch	Chum	10,000,000	50,000	200
Wally Norenbury	Chum	50,000,000	100,000	500
GRAND TOTAL	Chum	60,000,000	150,000	400
Ft. Richardson				
Whittier	Coho	100,000	20,000	5
Cordova	Coho	60,000	10,000	6
Solomon Gulch	Coho	1,000,000	50,000	20
Wally Norenburg	Coho	3,000,000	100,000	30
GRAND TOTAL	Coho	4,160,000	180,000	23
Main Bay	Sockeye	2,500,000	100,000	25
GRAND TOTAL	Sockeye	2,500,000	100,000	25
Wally Norenburg	King	150,000	50,000	3
GRAND TOTAL	King	150,000	50,000	3
GRAND TOTAL	ALL	691,810,000	1,521,667	455

Appendix A

Tagging goals

System	Treatment	Species	Projected Release	Valid Tag Goal	Total Release /Marked Ratio
	Oiled	Pink	1,000,000	50,000	20
	Oiled	Pink	1,000,000	50,000	20
	Oiled	Pink	1,000,000	50,000	20
	Clean	Pink	1,000,000	50,000	20
	Clean	Pink	1,000,000	50,000	20
	Clean	Pink	1,000,000	50,000	20
GRAND TOTAL	ALL	Pink	6,000,000	300,000	20
Coghill	Clean	Sockeye	1,000,000	50,000	20
Eshamy	Oiled	Sockeye	1,000,000	50,000	20
Jackpot	Oiled	Sockeye	200,000	50,000	4
GRAND TOTAL	ALL	Sockeye	2,200,000	150,000	15
GRAND TOTAL	ALL	ALL	8,200,000	450,000	18

CONFIDENTIAL

STATE/FEDERAL NATURAL RESOURCE DAMAGE ASSESSMENT
DETAILED STUDY PLAN

DRAFT

Project title:	Early marine salmon injury assessment in Prince William Sound
Study I D Number:	Fish/shellfish study No. 4
Lead Agency:	State of Alaska, ADF&G Fisheries Rehab, Enhancement and Development Division
Cooperating agencies:	Federal: NMFS Auke Bay Lab Univ. of Alaska: Inst. of Marine Science
Principal Investigator:	Jim Raymond, Fishery Biologist
Assisting personnel:	Alex Wertheimer, NMFS R. Ted Cooney, IMS
Date submitted:	11 October 1989

	Signature	Date
Principle Investigator:	Jim Raymond	10/11/89
Supervisor:		
Consulting Biometrician:		
OSIAR Senior Biometrician:		
OSIAR Program Manager:	Bill Hobson	10/13/89
OSIAR Director:		

Note: This study plan is for the period 3/1/89 - 2/28/90. It does not accurately describe plans for the 1990 season. It is expected that methods used in 1990 will be revised after data collected this year has been analyzed.

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OVERVIEW

INTRODUCTION

The early marine period is a critical one for salmon because it is at this time that the greatest mortality is sustained (Parker 1968; Bax 1983, Hartt 1980; Foerster 1968; Ricker 1976; Nichelson 1986). Mortality is considered to be inversely proportional to the rate of growth, since a prolonged juvenile period will result in a prolonged vulnerability to predators (Parker 1971; Healey 1982; Taylor 1977; Walters et al. 1978). For a possible exception to this, see Helle (1980). Therefore, factors that lower normal growth rates during the early marine period, such as toxic effects of exposure to hydrocarbons, reduction in prey populations, or increased energy expenditures associated with the disruption of normal migratory patterns, could have a strong influence on survival.

Oil in the marine environment can affect juvenile salmon in a variety of ways. Oil can be directly toxic to salmon; juvenile salmon are especially susceptible when first in seawater (Rice et al. 1975; Rice et al. 1984). Sublethal levels of hydrocarbons can affect metabolism and reduce growth of juvenile salmon (Rice et al. 1975). Sublethal levels of water-soluble hydrocarbons can also damage olfactory lamellar surfaces, conceivably impacting migratory behavior and feeding patterns (Babcock 1985). Oil can also be toxic to littoral and pelagic macroinvertebrates (Caldwell et al. 1977; Gundlach et al. 1983). Thus, mortality, reduction of reproductive potential, or growth inhibition of prey populations could reduce growth rates of juvenile salmon, and thus increase their exposure to predation.

During the past decade, five world-class hatcheries have been established within Prince William Sound. These facilities, operated by the Prince William Sound Aquaculture Association, Valdez Fisheries Development Association and the State of Alaska, produced approximately 535 million juvenile salmon in 1989. The hatchery contribution represents roughly half of the total number of juvenile salmon produced in PWS this year. A coded wire tagging program marked roughly 1.3 million juvenile salmon this year. Approximately one in every 1,000 juvenile salmon in PWS this year was expected to have a CWT. Recoveries of these marked fish in PWS will play a major role in our assessment of the impact of the oil spill.

Because salmon constitute the great majority of the economic resource of PWS, a major effort will be directed at early marine studies. The impact assessment will be conducted by the Alaska Department of Fish & Game, the National Marine Fisheries Service, and the Institute of Marine Science. The research will cover all phases of the early marine period in PWS salmon, from nearshore rearing to pelagic migration. Studies conducted by ADF&G will

focus on the impact of the oil on migratory behavior, studies conducted by NMFS will focus on pairwise comparisons of salmonid growth and behavior in oiled and un-oiled nearshore rearing habitats, and studies conducted by IMS will focus on a comparison of fry and plankton in an oiled site with data obtained in the same site a decade earlier (Cooney et al. 1981). Sampling will be coordinated to produce a single cohesive data bases of 1) coded-wire tag recoveries and 2) zooplankton and epibenthos collections with associated temperature data.

We have emphasized a coordinated approach to attaining the objectives. The studies are mainly complementary. A strong effort is required because of 1) the high value of the resource and 2) the wide range of habitats utilized by salmon during the early marine phase. The studies also supplement each other in areas where extensive sampling effort is required. In particular, strong efforts will be needed to recover tagged fish because of their relative rarity, and to assess the plankton biomass because of its patchiness. The three agencies will cooperate on the tag recoveries and plankton assessment.

GENERAL OBJECTIVES

The following objectives encompass the objectives of the three component studies in this project. Objective criteria for the objectives are described in the component studies below.

A. Estimate the effects of oil contamination on abundance, growth, feeding habits, and behavior of juvenile salmon during their early marine residence.

B. Estimate the effects of oil contamination on critical habitats utilized by juvenile salmon during their early marine residence.

C. Describe migration patterns of juvenile salmon relative to oiled and non-oiled areas of western Prince William Sound and the residence time of fish in the oil-impacted areas.

D. Estimate hydrocarbon levels in tissues of juvenile salmon in oiled and non-oiled areas and document oil-related mortalities.

E. Identify potential methods for restoration of lost populations, or habitat where injury is identified.

GENERAL METHODS

A. Approach

Objectives A,B. Three basic approaches will be used to assess effects of oil on juvenile salmon and their habitats: 1) Direct pairwise comparisons of oiled and non-oiled habitats and the abundance, growth, feeding habits, and behavior of juvenile

salmon in these habitats; 2) Comparisons with historical data on juvenile salmon ecology in Prince William Sound; 3) Differences in abundance, growth, feeding habits, and food resources of juvenile salmon relative to the geographical distribution of the oil contamination.

The study team realizes that measuring growth rates is a difficult task. In the case of untagged fish, the populations being sampled at different times and different places will, in general, be different. Another problem that will affect growth measurements of both tagged and untagged fish is size-selective mortality: the growth that is measured will actually be the growth of the survivors. To reduce these problems, conclusions concerning growth of juvenile salmon will not be based on size alone. They will be based on several growth-related factors including size, condition factor, stomach fullness, food abundance and temperature.

Objective C. The impact of the oil spill on juvenile salmon migration patterns will be determined primarily by the recoveries of coded-wire tags from juvenile salmon released from PWS hatcheries. The tags will provide information on the speed, route, and residence times of specific labeled groups. Tag recovery rates will also indicate relative abundance of wild and hatchery fish at sites throughout western PWS. Catch per unit effort data will be used to determine the relative utilization of different migration corridors.

Objective D. The level of hydrocarbon contamination of the tissues of juvenile salmon will be assessed by pairwise comparisons between oiled and non-oiled areas. Coded-wire tagged salmon will be collected in such a manner that fish of known point of origin can be analyzed for hydrocarbons if sufficient recoveries of specific tag lots are made in a particular time and area.

Objective E. The study leaders will stay alert to possibilities for the restoration of habitat and populations that may be lost or damaged by the oil spill.

B. Standard operating procedures

Unless otherwise described, sampling and sample handling procedures were standardized as follows:

Zooplankton was sampled with 20-m vertical tows using 216 and 243 u mesh, 0.5 m-diameter nets. Horizontal tows were not done due to the presence of oil in many of the sampling sites. Epibenthic invertebrates were sampled with 10-m horizontal tows using 243 u mesh, 0.3-m-diameter nets at depths of 0.5 m during tide stages between -1 and +3 ft.

Beach seine specifications were as follows: 37-m-long; outer wings, 10 m long, 32 mm "scare" mesh, dyed green, tapering from 1 to 3 m deep; inner wings, 4 m long 13 mm mesh dyed green, tapering from 3 to 4 m deep; bunt, 8 m long, 6 mm mesh dyed green, tapering from 4 to 5 m deep.

To avoid excessive mortality when large numbers of fish were caught, fish were placed in a holding tank until processing was completed. Lots of approximately 300 ml of fish (measured by displacement in a 1-liter beaker) were put through a 2-inch tunnel tag detector (Northwest Marine Technologies) with a small stream of salt water. When a tag was found in the lot, the lot was continuously divided until the tagged fish was found. One 300-ml sample of fry was sorted immediately to determine species composition and released. Another sample of approximately 80 fry was preserved in buffered formalin for later size measurements and stomach analyses. Remaining fry were released. All tagged fish were blot-dried, weighed and measured (snout to fork). The tagged fish were then assigned an ID number and frozen individually. Tags on these fish will be read later by the FRED Tag Lab. There the heads will be excised from the frozen fish, the body returned to the original sample vial, and the head assigned a tag lab head number corresponding to the original sample number. The frozen bodies will then be returned for storage to the Auke Bay Laboratory until priority for hydrocarbon analysis is determined. Chain of Custody procedures will be used throughout transfer and storage of these samples. After the tags are read, an attempt will be made to pool the fish in 10-g lots for the hydrocarbon analysis. Excess fish will be retained for stomach analyses. Untagged fish were left in buffered 10% formalin for at least 30 days to standardize shrinkage. These fry were rinsed in buffered sea water, blot-dried, weighed and measured. Moribund fish, if found, were to be placed immediately in Bouin's solution for later histopathology studies.

Currents were measured with Marsh-McBirney model 201 current meters at depths of 0.5 m. Water temperatures were measured at 0.5 and 5.0 m depths.

I. Impacts of oil spill on migratory behavior, growth and mortality.

Project leader: Jim Raymond, ADF&G

Determination of the impact of the oil spill on the migratory behavior and growth of salmon juveniles in PWS in 1989 will largely be based on the recovery of tagged salmon that were released at various points in the Sound from late April through early June.

OBJECTIVES

(Letters refer to General Objectives, above)

C-1. Document the impact of oil on the migratory path, speed of migration, and residence time of CW-tagged salmon releases in PWS. Determine at the alpha=.05 level whether speeds and residence times are different in oiled and unoiled areas and in oiled and unoiled years.

A-1. Compare the growth related factors between CW-tagged salmon captured in oiled and un-oiled areas. Determine at the alpha=.05 level whether size, condition factor and stomach fullness are different in CW-tagged fry collected in oiled and unoiled areas and in oiled and unoiled years.

D-1. Document the impact of oil on the hydrocarbon content of CW-tagged fry having known residence times. Determine at the alpha=.05 level whether hydrocarbon content differs in CW-tagged fry collected in oiled and unoiled areas.

D-2. Document the occurrence of dead and moribund fish, and collect tissues from these fish for hydrocarbon and histopathology analyses.

METHODS

A. Study design

Tag lots were released during time periods ranging from 1 to 30 days (10-day average). Recovery of these salmon at later times and in different places will allow estimates to be made of migration paths, migration speeds and residence times, and relatively accurate measurements of growth. Residence times may also provide information on food abundance and growth, since salmon juveniles tend to move out of areas with low food abundance. Approximately 1.3 million tagged fish were released. The untagged/tagged ratio ranged from 289 to 924 for each tag lot (500 average). It is presently assumed that the wild and hatchery populations are about equal, indicating that collection of approximately 1,000 salmon will be required to recover one tag.

Thus, an effort was made to recover several thousand juveniles per day.

To measure the impact of the oil spill, two types of comparisons will be used. First, juveniles collected at un-oiled sites and having known residence times in the Sound will be compared with juveniles collected at oiled sites having similar residence times. Second, juveniles collected in oiled areas in 1989 will be compared with juveniles collected in the same areas in succeeding years of this project when lower pollution levels are expected. Data obtained on the untagged fish will serve as a second, although less accurate, source of information on the effect of the oil on growth, distribution and tissue pollutant levels. The impact of the oil spill on mortality will be based on observations of dead and moribund fish, and hydrocarbon and histopathological analyses of selected tissues.

B. Collection areas

Six sample collection areas (Fig. 1) were selected specifically to recover CWT salmon juveniles released from five hatcheries and two streams:

1. North Perry Island area. This site is expected to yield collections of fry and smolts from VFDA, Cannery Creek and Esther hatcheries and wild sockeye smolts tagged at Coghill Bay. It appears that this site will remain un-oiled.

2. Crafton Island - Pt. Nowell. This area is expected to yield collections of releases from the above sites, Main Bay Hatchery and the wild sockeye tagged in Eshamy Bay. Oil fouled the Pt. Nowell area in late April.

3. Chenega Eddy between Whale Bay and Pt. Countess. This area has been previously identified as a fry holding area and is expected to yield collections of tagged fish from all the above releases. It is a heavily oiled site.

4. North Twin Bay area. This area is expected to yield collections of tagged fish from all the above sites. Collections in these areas will determine the distributions of these fish in the Elrington, Prince of Wales and Bainbridge passages. It is a heavily oiled site.

5. Sleepy Bay - Pt. Helen area. This area will allow the measurement of the relative importance of (1) Montague Strait and LaTouche passages and (2) Knight Passage and lower Montague Strait as migration routes for the above releases. It is a moderately oiled area.

6. Hinchinbrook Entrance. Collections in this area will help determine whether tagged salmon use this passage to leave

the Sound. It is thought that in normal years, few hatchery salmon leave the Sound by this route. However, because it is an un-oiled area, some tagged fish might choose this route this year.

These areas were sampled approximately once a week from 5 May through 11 July. The site at the Hinchinbrook Entrance was sampled approximately bi-weekly due to the lower tag recoveries expected there. More frequent sampling was to be done at this site if a significant number of tags are recovered. One day was spent at each area. When time was available, additional sites that appeared to be along important migration pathways were included.

C. Logistics

A 50' seiner equipped for night navigation was chartered for the period 8-22 May. From 25 May to 11 July, the Sundance was used. A crew of three Fish & Game personnel operated the sampling gear and skiff. At each sampling area, sampling was conducted approximately between 1000 and 2230 h. The boat then moved to the next station and moored.

D. Sampling procedures

1. Beach seine and dip nets. Most collections, especially early in the season, were collected with a beach seine (described above) and 18" diameter dip nets on two or three beaches in each sampling area.

2. Tow net and purse seine. Two replicate offshore samples were obtained with 10-min tows of a standard FRI tow net pulled between two boats. The tow net was operated after dark to increase its efficiency. A 200 ft purse seine (Auke Bay design) was used in late June and July. It was used in areas where concentrations of salmon juveniles were observed and where a beach seine could not be used.

3. Epibenthos samples. Samples were taken at inshore sites as described above. Volume and composition of selected samples will be determined at the FRED Limnology Laboratory. A maximum of one beach seine site a day (depending on tide height) was sampled with a single tow of the epibenthic plankton sled (described above). Samples were collected during daylight hours.

4. Oceanographic measurements. Water temperatures and salinity were measured with a YSI salinometer at depths described above. Surface current measurements were deleted from the study plan because initial measurements were uniformly zero in the sampling areas. Tide levels and directions were

recorded for each sampling site. These data will be used to help determine migration patterns of the salmon.

E. Fish handling procedures.

Tagged and untagged juvenile salmon were handled as described above. Approximately 60 untagged fish of a given species were randomly selected each day from both beach seine and tow net catches and preserved in formalin for later weight, length and stomach fullness measurements. This number should be sufficient to identify different size groups if they should occur. Stomach analyses (for 10-fish samples) will be done at the FRED Limnology Laboratory.

DATA ANALYSIS

In each of the following comparisons, except where noted, the hypothesis that there is no difference between CW-tagged pink fry collected in oiled and unoiled areas will be tested with t-tests at the alpha = 0.05 level. For correlations, the hypothesis that r=0 will be evaluated at the alpha = 0.05 level.

A. Growth data

Because many factors other than the presence of oil can influence growth, several analyses of growth-related factors will be done to help isolate the impact of oil:

1. Comparison of oiled and un-oiled areas in 1989.
   a. growth curves expressed as percent increase in length and weight per day
      i. CW-tagged fish using size data at time of release.
      ii. Untagged fish using fish caught at same site at different times in the season (apparent growth rate).
   b. condition factor at representative times; tagged and untagged fish.
   c. stomach fullness; primarily untagged fish. (sufficient tagged fish not expected to be available)
   d. epibenthos biomass density.

2. Comparison of oiled areas of 1989 with same areas in following years using growth-related factors shown in 1a-d.

3. Correlation of hydrocarbon levels with growth in 1989; tagged fish.

4. Correlation of epibenthos biomass and growth in 1989.

B. Migratory speeds and patterns

Migratory speeds will be calculated using the minimum distance between release and recovery sites and the average release date for a given tag lot.

The best method for describing the migratory patterns will not be apparent until the migratory data is available. A method currently under consideration is the construction of "migratory roses" (similar to wind roses used by meteorologists). These would be centered at hatchery locations and consist of arrows to each recovery site. Specific analyses will include:

1. Comparison of migration of CW-tagged pink fry in oiled and un-oiled areas in 1989. The unoiled areas will include recovery sites of fry released from VFDA, Cannery Creek and Esther hatcheries and the oiled areas will include recovery sites of fry released from AFK Hatchery.
   a. Migratory speeds.
   b. Migration patterns (e.g., migratory roses).

2. Comparison of oiled and unoiled areas of 1989 with same areas in following years.
   a. Migratory speeds.
   b. Migration patterns.

SCHEDULE/PERSONNEL

	1989										1990	
Activity	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec	Jan	Feb
Sampling		xxxxxxxxxxxx										
Data compil.			xxxxxxxxxxxxxxxxxxxx									
Data analysis							xxxxxxxxxxxxxxxxxxxxxxxxxxxxxxxx					
1990 season plannng							xxxxxxxxxxxxxxxxxxxxxxxxxxxx					
Project coordination						xxxxxxxxxxxxxxxxxxxxxxxxxxxxxxxxxxxx						

Personnel

J. Raymond	Project leader 1 Apr-28 Feb
M. Willette	Field crew leader 5 May-13 July
FTII (2 posns)	Operate sampling gear 5 May-11 July
FTII	Measurement of salmon fry 1 Aug-1 Sep
FTIII	Coded wire tag reading (Tag Lab employee)

BUDGET 3/1/89 - 2/28/90

100

Name	Class	Months	Salary	total	
Jim Raymond	FBIII	10.0	6.0	60.0	
Mark Willette	(RSA)	3.5	4.5	15.8	
Dave Morris	FTII	2.5	5.0	12.5	
Scot Fiscus	FTII	2.5	5.0	12.5	
Marlene Luke	FTII	1.0	3.0	3.0	
David Petree	FTIII	1.0	4.0	4.0	
				Total	107.8

200

3 RT Fairbanks/Cordova @344	1.0	
2 RT Fairbanks/Anchorage @227	0.45	
2 RT Fairbanks/Juneau @346	0.7	
Per diem 15 @ $90	1.35	
	Total	3.5

300

Vessel charter "Kittiwake"	28.0	
Food and fuel	3.0	
Vessel use "Sundance" food, fuel	5.0	
plankton analyses 15 @ $100	1.5	
stomach analyses 50 @ $100	5.0	
tag lab CWT reading	3.0	
telephone 8 mo @ 300	2.4	
air & helo charter, PWS	1.3	
air freight equipment	0.4	
equipment repair	0.8	
office expenses, misc.	0.2	
	Total	51.1

400

tow net	1.0	
beach seines 2 @ $900	1.8	
dip nets	0.3	
purse seine	1.0	
plankton nets	0.5	
sample bottles	0.6	
chemicals	0.3	
survival gear, rain gear	2.0	
small boat fuel 250 gal @ 1.20	0.3	
spare propellers 2 @ 100	0.2	
misc. small boat parts	0.5	
misc. scientific supplies	1.2	
	Total	9.7

500

18-ft skiff & trailer, 90 hp OB	12.0	
75 hp OB	3.0	
CWT OMNI detector	7.5	
March McSirney current meter	2.2	
freezer, 5 cu ft	0.6	
	Total	25.3
	GRAND TOTAL	197.4

II. Impact of Oil Spill on Juvenile Pink and Chum Salmon and their Prey in Critical Nearshore Habitats

Project Leader: Alex Wertheimer, NOAA, NMFS
Auke Bay Fisheries Laboratory

The NMFS component of the early marine salmon studies will focus on comparisons, between oiled and non-oiled areas of Prince William Sound, of salmon distribution, abundance, size and nominal growth rates, feeding habits, and prey abundance. These parameters will be examined quantitatively to assess if a significant impact occurred to the juvenile salmon or their food resources.

OBJECTIVES

(Letters refer to General Objectives, above).

A-1. To test at alpha = 0.05 the hypothesis that the abundance of juvenile pink and chum salmon does not differ between oiled and non-oiled areas.

A-2. To test at alpha = 0.05 the hypothesis that the size and growth rates of juvenile salmon does not differ between oiled and non-oiled areas.

B-1. To test at alpha = 0.05 the hypothesis that the prey available to juvenile pink and chum salmon in the littoral and pelagic water column does not differ between oiled and non-oiled areas.

A-3. To quantify the feeding habits of juvenile pink and chum salmon in terms of frequency of occurrence, biomass, and Index of Relative Importance, and to compare oiled and non-oiled areas using Percent Similarities Index.

A-4. To test at alpha = 0.05 the hypothesis that indices of stomach fullness do not differ between oiled and non-oiled areas.

C-1. To determine migratory behavior of juvenile salmon based on coded-wire tag recoveries; tag recoveries will be incorporated in the cooperative data base managed by ADFG.

D-1. To test if hydrocarbon levels in juvenile pink salmon differs between oiled and non-oiled areas.

METHODS

A. Sample Design

Nearshore habitats will be sampled according to a 2x2x3 factorial design. The first level refers to oiled and non-oiled areas of western Prince William Sound. The second level refers to two

general classifications: embayments and migration corridors. For both oiled and non-oiled areas, two each embayment and corridor locations will be selected, for a total of 8 locations. The third level (sites) refers to habitat types, grossly characterized by gradient and substrate: low gradient beach (<10% grade, granule-pebble substrate); medium gradient beach (12-25% grade, pebble-cobble substrate); and steep gradient beach (>50% grade, bedrock or large boulder substrate). Particular sample beaches will be selected for similarity between oiled and non-oiled areas in such characteristics as wave exposure, macrophyte coverage, and substrate. A total of 24 sites will be regularly sampled. If sample days are lost on a trip due to weather, embayment sampling will have priority over corridors.

Biological sampling will extend from mid-April to late June. A total of 5 sampling cruises of approximately 2 week duration will be required with a 2 day interval between cruises. Each cruise will be comprised of two separate sampling components.

The first component will be sampling at the 8 locations during the -1 to +3 ft tide range to collect fish samples for relative abundance, size, growth, feeding habits, and coded-wire tag (CWT) analysis (Figure 2). Environmental data, pelagic zooplankton, and epibenthic zooplankton will be collected at each site. Sampling will alternate daily between oiled and non-oiled areas in western Prince William Sound. Matched pairs of oiled and non-oiled sites will be chosen so that the pattern of alternation between areas is maintained for the duration of the study. This sampling scheme will allow direct pairwise comparisons between oiled and non-oiled locations.

The second component will include further sampling at the 4 embayments for epibenthic macroinvertebrates at 2 ft intervals from the -1 to +9 ft tide level. This sampling will provide a measure of variance of epibenthic prey within the -1 to 3 ft tide window in addition to supplementing the epibenthic zooplankton sampling in component 1 to provide a weekly time series on epibenthic prey at the embayments. This sampling will also provide a measure of epibenthic prey fields at the higher tide levels (>+3 ft) typically more heavily impacted by oil deposition.

B. Description of Study Sites

Grade for each sample site will be measured using a hand-held level and stadia rod, and determining elevation change over a measured distance (10-15m). Substrate will be characterized at the +1 tide level. At each end and at the middle of a the sample area, a 0.25 m^2 area will be assessed as to percent of surface coverage by different sized substrates. The Wentworth scale (Holme and McIntyre 1984) will be used to classify materials. Particles smaller than granule (2 mm) will be classified as sand. The mean

value for the three quadrats will be used to characterize the beach.

Macrophyte coverage will be estimated by walking the length of the sample area at water depths of 0.3 and 0.6 m below given tidal heights. Percent coverage will then be estimated for kelp, eelgrass, filamentous algae, and Fucus.

In addition to the above levels of characterization of substrate and macrophyte coverage, photographs of 0.1 m^2 quadrats will be taken at 2-ft elevation intervals at the embayment sites, from the -1 to +9 ft tide level. Six quadrat locations will be selected randomly along an 18 m tape stretched parallel to the water line. The meter tape will be included in the photograph for scale. The photographs will be subsequently evaluated for substrate composition and percent macrophyte coverage.

The general area within 1 km of the sample sites will also be characterized as to grade and substrate in the -1 to +3 ft tidal range to determine the relative proportion of the different habitats within each location. Because of the time constraints of the biological sampling during the April-June period, this habitat classification will be done on a separate cruise in July.

C. Sample Collection

1. Fish Samples

Sampling at the study sites will be restricted to the -1 to +3 tide levels to standardize tidal effects between sites. Fish will be captured using 37 m beach seines (described above) and a seine modified to sample the cliff sites. The cliff seine is 37 m long and 3 m deep. The wings are 10 m long with 32 mm "scare" mesh dyed white. The bunt is 17 m long with 6 mm mesh dyed green and has a floor of 6 mm green mesh formed by a 9 m lead line connecting the bottom intersections of the wings wiht the bunt.

Catches will be sorted to species and enumerated; all salmon will be checked for the presence of coded-wire tags using an OMNI coded-wire tag detector. Each coded-wire tag salmon will be measured fresh for fork length and weight and frozen in separate 20-ml glass vials. On each sampling trip, up to 30 each juvenile pink and chum salmon from each sample site will be preserved in formalin for later length and weight measurements; 10 of these fish will also be used for diet analysis. The fish will be placed in oil sample bags, with up to 15 individuals of a species per bag. Each bag is labeled as to date, set number, time, site, habitat, and number of fish. The bags will be stored in a 3.8 liter jar containing 2 liter 10% formalin; one jar is used for the collections from the three habitats at a particular site for each sampling date.

In addition, up to three replicate samples of 50 juvenile pink salmon will be retained for hydrocarbon analysis, as per standard operating procedures. These fish will be placed in 120-ml jars while alive and frozen. All other fish will be released.

As time permits, the shoreline within the general vicinity of the habitat sites will be surveyed to locate congregations of juvenile salmon. Beaches will be sampled with beach seines by both "blind" sets (no fish observed) and "directed" sets (fish observed). Schools of juvenile salmon will also be sampled using dip nets where seine sets are not feasible. These collections of juvenile salmon will be enumerated, checked for coded-wire tags, and used to supplement collections for hydrocarbon and stomach analyses when insufficient numbers are collected at the regularly sampled beaches. All other fishes caught in such sets will also be identified and enumerated.

2. Prey Abundance

Potential prey of the juvenile salmon will be sampled by epibenthic sled and vertical plankton hauls. At each of the three sites within each location, the 0.5 m water depth (relative to tide height at time of sampling) will be sampled for macroinvertebrates with an epibenthic sled carrying a 0.3 m diameter 243 micron net. A measured 10-m horizontal haul will be made. In the offshore water adjacent to the habitats sampled, triplicate samples of pelagic zooplankton will be taken with a 20-m vertical haul of a 0.5 m diameter 243 micron net. Water of sufficient depth for the haul will be located using a depth-sounder or a sounding line.

A series of epibenthic sled samples will be taken on the lowest tides of each trip at the embayment sites as part of the component 2 sampling (see Section A above). The three sites at each embayment location will be sampled at 2 ft tide intervals from -1.0 to +9.0 ft tide levels.

Preservation of the samples will be similar for both epibenthic and pelagic samples. A battery-powered seawater pump will be used to wash down the outside of the net. The contents of the cod-end are then rinsed into a 500-ml plastic bottle containing 25-ml concentrated formaldehyde and enough filtered seawater to fill the bottle 1/2-2/3 full. The cod-end of the plankton net will be rinsed using a plastic squirt bottle and filtered seawater. The sample bottle is filled to the top, so as to make a 5% formalin solution. The bottle will be labeled as to site, date, gear, and habitat. Both an internal and external label will be used. Sample bottles will be sealed by using plastic thread-sealant tape inside the lid, and plastic electrical tape on the outside of the lid, to prevent spillage or leakage.

3. Hydrocarbon samples

In addition to the samples of juvenile pink salmon mentioned previously, water samples, mussel samples, and sediment samples will be taken for hydrocarbon analysis at each location. Two replicate samples of water will be taken at 1.0 m and 4.0 m offshore of each site on each sampling date. The hydrocarbons will be extracted immediately using dichloromethane and the extracts frozen or kept on ice. Mussels will be sampled at or near one of the habitats within each site, and kept on ice or frozen. A 480-ml glass jar will be filled as a sample for each site on each sampling date. Sediments will be sampled at the water line at each habitat within a site; a 120-ml glass jar will be filled from several randomly selected spots within the beach seine area, using a solvent-clean spoon. During the epibenthic sled sampling of the -1 to +9 ft tidal range at the embayment sites, a sediment sample will be taken in association with each sled tow. All sample jars and collecting equipment will be solvent-cleaned or heat-cleaned. See Auke Bay Laboratory Water Quality and Sediment Study Plans for more detailed protocol.

4. Environmental data

Water temperature and salinity at 0.5 m depth, wave height, and current measurements will be taken at each nearshore habitat regularly sampled. Water temperatures at 1 m and 4 m will be taken in association with each set of zooplankton tows and water samples. Temperature and salinity will be measured using a Beckman probe conductivity-temperature meter. Current will be measured with a Marsh-McBirney induction current meter. Wave height will be measured with a meter stick. Extent of oil deposition and of visible oil in the water will also be noted for each habitat.

E. Sample Processing

1. Fish Samples

a. Coded-wire tagged fish

Coded-wire tagged fish will be stored frozen until processing for hydrocarbon content. The fish will be transported from field collection to the Auke Bay Laboratory, then to the ADFG Tag Processing Laboratory in Juneau. There the heads will be excised from the frozen fish, the body returned to the original sample vial, and the head assigned a tag lab head number corresponding to the original sample number. The frozen bodies will then be returned for storage to the Auke Bay Laboratory until priority for hydrocarbon analysis is determined. Chain

of Custody procedures will be used throughout transfer and storage of these samples.

The tag lab personnel will decode the tags, and transmit the information to NMFS and to the ADFG investigator coordinating Early Marine Salmon Studies.

b. Size and stomach samples.

Samples will be stored in formalin at least six weeks before processing to standardize shrinking. Before processing, samples will be soaked in freshwater for 1-3 hr to reduce formalin fumes. The fish from a particular set will be arranged on a tray by species, and up to 10 of each species per set selected randomly for stomach analysis and given an appropriate individual sample number. The fish will then be measured to the nearest mm fork length, then blotted lightly on a paper towel and weighed to the nearest mg. Hard copies of the data, along with corresponding sample information, will be retained.

After being weighed and measured, each fish retained for stomach analysis will be put into a labeled 20-ml vial filled with 50% isopropyl alcohol or 70% ethanol. Subsequent analysis will involve excision of the foregut and estimation of stomach fullness. The foregut will then be weighed, the contents removed, and the empty foregut reweighed to give a measure of total content wet weight. The prey items will be identified to a minimum of Order level and counted. Biomass of prey taxa will be estimated from dry weights computed for the same or similar taxa in other feeding habits research (Landingham 1982; Landingham and Mothershead 1988), or computed from intact prey if adequate literature values are not available by weighing a sample of 100 intact representatives of a taxa dried in a constant-temperature oven at 60°C for 24 hr.

2. Zooplankton and epibenthic samples

Upon transport to the Auke Bay Laboratory, the samples will be logged in by sample number. Contracts will be put to bid for analysis, with competitive bid procedures used to select among qualified applicants. For each sample, the contractor will provide hard copies and ASCII files on floppy disk with the following information:

Sample number, volume, and wet weight
For each organism:
name, to genus for harpacticoid and calanoid copepods, all others to order

Life History Stage
Number per cubic meter (total, mean, range, sd)
Wet weight (gm/m^3), average biomass (mean,sd)
Percent abundance and biomass
Total number of categories
Shannon-Weiner and Brillouin diversity indexes (Pielou 1975)

Because epibenthic samples often contain a considerable amount of plant material, some samples will be precleaned at the Auke Bay Laboratory before they are sent to the contractor. Precleaning will involve picking all animals and eggs from the plant debris. The plant debris will be saved, and will be checked by a staff biologist (other than the person who cleaned the sample) prior to the sample being shipped to the contractor, to ensure the sample has been picked clean. When samples are sent to the contractors for analysis each sample number will be recorded and checked against the original sample catalog. All samples are shipped via the U. S. Postal Service, certified mail, return receipt requested. All shipping documents are retained at the Auke Bay Laboratory.

3. Hydrocarbon samples.

All hydrocarbon samples (fish tissue, water, sediments, and mussels) collected in the course of this study will be prioritized by the Hydrocarbon Analysis project as to if and when the samples will be processed. Procedure for analysis of these samples is detailed in the Hydrocarbon Analysis study plan.

F. Data Management

As much as possible, field and laboratory collection data will be entered on forms for fish sample collections, including associated environmental data; coded-wire tag collections; environmental data for epibenthic sled tidal series collections; length and weight measurements; and stomach sample analysis. All other associated data will be recorded in field and laboratory notebooks cataloged as described below in the data archival system. Data will be entered in as timely a fashion as possible into the appropriate computer data bases. R-Base/Dos on microcomputers will be used as a data base system. All data entered will be verified by checking computer-generated hard copies against the original data records.

DATA ANALYSIS

Preliminary data analysis will involve tabulation of data and graphical comparisons of parameter values between oiled and non-oiled sample sites. The following factors will be tested using analysis of variance (ANOVA): abundance and size of juvenile salmon; prey numbers and biomass (prey samples); and stomach

fullness, prey numbers, and prey biomass (gut samples). Whenever an ANOVA F-test for simultaneously comparing several parameter means is found to be statistically significant, differences between means will be tested using appropriate multiple comparison procedures such as the Neuman-Keulls test (Winer 1971). It is recognized that empty cells may preclude complete ANOVA tests, and alternate parametric or non-parametric tests may need to be applied (Milliken and Johnson 1984). Nominal growth rates between oiled and non-oiled areas will be compared using a exponential growth model, and comparing the regression slopes of Ln weight over time with analysis of covariance (Zar 1974). Percent similarity indexes between oiled and non-oiled areas will be developed for both prey and gut samples (Whittaker 1952).

SCHEDULES AND PLANNING

A. Data Submission Schedule

The table below summarizes milestone dates and activities. Other than the reports prepared for the Management Team, the only special report will be a listing of the tag code and associated sample data provided to the ADFG Principle Investigator, Early Marine Salmon Studies. Formal publication of data analysis will be precluded until release by the Management Team.

MILESTONE CHART.. x = Planned completion date
\- = Actual completion date

	1989									1990		
MAJOR MILESTONES	A	M	J	J	A	S	O	N	D	J	F	M
Field activity, cruise reports	-	-	-	-								
Completion of epibenthic sampling processing (-1 to +3 tide)						X						
Epibenthic sampling processing (+5 to +9 tide)											X	
Zooplankton sample processing							X					
Size data (juvenile salmon)						X						
Stomach sample processing											X	
Data Analysis, Abundance, Size, Growth							X					
Data Analysis, Prey Fields							X					
Data Analysis, Feeding Habits												
Reports: Abundance, size, growth, prey fields											X	
Reports: Stomach analysis												

B. Data and Sample Archival

All field and laboratory data forms generated through the course of this study will be placed in notebooks numbered according to the Auke Bay Laboratory Oil Spill Notebook Tracking System (NTS). All field notes will be similarly cataloged. Trip reports for each sampling cruise and study plans will also be archived within the NTS. Copies of computer data files will be maintained on two microcomputer hard drives, as well as rotating floppy disk back-up kept in a locked cabinet.

C. Management Plan

Project will be managed by the project leader, under the general direction of the organization leader. Budget expenditures will be tracked using the NMFS Auke Bay Laboratory budget management system. All procurement, contracting, and labor hires will also be according to Department of Commerce, NOAA, procedures, and be undertaken through the procurement and personnel office at the NMFS Auke Bay Laboratory.

The following is a list of scientific personnel associated with the project. The individuals listed are permanent staff biologists at the Auke Bay Laboratory assigned on a part-time basis to the project, except for Gish and McGregor, hired specifically for the study.

Alex Wertheimer, Project Leader, Chief Scientist on sampling cruises
Adrian Celewycz, Alternate Chief Scientist
Mark Carls, Hydrocarbon Sampling, Water Chemistry
Joseph Orsi, Sampling and logistics
Don Mortensen, Sampling and logistics
Herbert Jaenicke, Sampling and Logistics
Robert Gish, Sampling and Stomach sample analysis
Susan McGregor, Stomach sample analysis
Joyce Landingham, Supervision of lab sample processing; contracting official technical representative for zooplankton and epibenthic sample contracts
Molly Sturdevant, coordination of lab sample processing

D. Logistics

Sampling equipment, including large gear such as nets and motors, will be transported from Juneau to Cordova using NOS vessels as available. Initial response will require use of scheduled air freight and Air National Guard C-130 assistance.

Sampling will occur in the western portion of Prince William Sound (see attached maps for study sites). A vessel of sufficient size to support a scientific crew of 4-5 and to transport sampling gear and two 17-ft skiffs is required. NOS

vessels will be used as available. Cruises will originate in Cordova and last approximately 14 d, including transit time from Cordova to study area. A total of six sampling cruises will be required.

Personnel and sample transport will principally be utilizing scheduled aircraft between Juneau and Cordova. In addition, charter float planes will be required for initial surveys of the study area, and to transport personnel and materials on an as-needed basis.

BUDGET

The budget for this project is shown in the following table. It should be noted that the labor costs shown do not include permanent salaries, except when permanent staff will be on duty in Prince William Sound. All permanent labor costs associated with planning, logistic support, sample processing, data analysis, and reports will be absorbed by existing project funds. Costs shown extend through February 1990.

100	LABOR		95 K
200	TRAVEL (Includes air charter)		13 K
300	CONTRACTUAL SERVICES		70 K
400	SUPPLIES AND MATERIALS	30 K	
500	EQUIPMENT	18 K	
TOTAL			226 K

III. Early Marine Salmon Studies in Prince William Sound and at the Armin F. Koernig Salmon Hatchery at Port San Juan, Alaska

R.Ted Cooney
Institute of Marine Science
University of Alaska Fairbanks

Studies at the AFK hatchery and from the R/V Alpha Helix are designed to quantitatively characterize salmon fry forage fields, fry feeding, small-scale migratory behavior and fry growth rates during the period April through June, 1989. Historical data bases at AFK (Cooney et al. 1978, 1981; Urquhart 1979; Barnard 1981) provide one means to determine whether previously established feeding dependencies by hatchery-released fry on local near-shore environments are being compromised by the presence of oil floating on the water or occurring on or in the beaches near Sawmill Bay. Shipboard studies of open Sound zooplankton establish source levels of forage populations advected into critical near-shore regions by tidal, wind and density driven circulation. Previous studies (Fulton 1983) in the adjacent Gulf of Alaska demonstrate that upper-layer zooplankton stocks washed into the Sound via the coastal current can vary by as much as a factor of 5.0 from year to year.

OBJECTIVES

The intent of University of Alaska studies is to assess Sound-wide forage stocks for wild and hatchery reared salmon fry prior to and during the crucial period of near-shore early ocean residence, and to procure a sample base for comparison with historical data available on zooplankton and fry feeding and growth at the AFK Hatchery at Sawmill Bay. Specifically the objectives are:

(Letters refer to General Objectives, above).

B-1. Determine the abundance of epipelagic zooplankton populations at specific locations in open Prince William Sound using a sampling design that estimates levels of variability associated with sampling a water column, with location and with cruise (April, May and June). Specifically, test the hypothesis that plankton populations are the same in oiled and non-oiled areas during all cruises. The design is capable of detecting differences of at least a factor of 2.0 with alpha=0.05.

B-2. Determine the abundance and levels of variability in forage stocks available to fry residing near the AFK hatchery at Sawmill Bay. Specifically, test the hypothesis that forage populations at oiled (outside the Bay) and non-oiled (inside the Bay) sites are the same using a sampling design capable of detecting weekly differences of at least a factor of 3.0 with alpha=0.05.

A-1. Determine the kinds, amounts and levels of variability of invertebrate food items in the stomachs of fry residing in and adjacent to Sawmill Bay. Specifically, test the hypothesis that food is the same for fish residing in (non-oiled) and outside (oiled) Sawmill Bay. The design is capable of detecting weekly differences of at least a factor of 2.5 with alpha=0.05.

A-2. Determine the rates of growth for hatchery-released fry residing in the near-shore zone of Sawmill Bay and adjacent Elrington Passage for comparison with historical data and with similar measurements being made by ADF&G and the NMFS at other locations in Prince William Sound. Specifically, test the hypothesis that fry growth rates near AFK are the same in oiled and non-oiled nursery areas. The design is expected to yield results capable of detecting growth rate differences of at least 1.0 percent of the body weight per day with alpha=0.05.

C-1. Determine the use patterns and small-scale migratory behavior of fry released from hatchery net pens in Sawmill Bay for comparison with AFK historical data.

D-1. Identify and count all dead and moribund fishes encountered in the process of sampling fry and their food in and near Sawmill Bay.

D-2. Scan total catches of fry (dip net and beach seine) for coded wire tags and recover and store tagged fry (frozen) for ADF&G growth and migration studies and NMFS hydrocarbon measurements.

METHODS

1. Fry forage stocks: Samples of invertebrate populations serving as food for fry will be obtained at selected locations in the main body of Prince William Sound and at historical sites in and adjacent to Sawmill Bay (hydrographic stations 1 and 4). In Prince William Sound proper, samples of epipelagic zooplankton will be obtained in duplicate 1-m net tows (0.505-mm Nitex) fished vertically through the upper 100 m or from 5 m above a shallower seabed. Sampling locations will include oiled (western) and non-oiled (eastern) areas. Zooplankters will also be collected in duplicate 20-m vertical tows (1/2-m net; 0.333-mm Nitex) at historical stations 1 and 4 near the AFK hatchery, and in replicated and metered (General Oceanics calibrated flowmeter) horizontal tows accompanied fry sampling at selected locations.

   All plankton samples will be preserved immediately in 10% seawater formalin for analysis at the hatchery or later at the University of Alaska Fairbanks.

Numbers and kinds of zooplankters in preserved samples will be estimated by standard subsampling techniques in which 100-150 animals are removed quantitatively from each collection using a calibrated pipette. Specimens are identified to the lowest taxon possible (generally species, sometimes genera, occasionally family or order) and tallied on standard laboratory count sheets. Data are reported as numbers m-3. A portion of each open Sound sample will be dried for 24 hr. at 60 deg C as a quantitative measure of standing stock.

2. Fry size and growth: Fry will be sampled using a 150- ft. beach seine and/or dip nets at historic and other selected sites in and near Sawmill Bay. Fry sampling will be conducted twice or more often each week (weather permitting). 200 fry will be saved from each seine or dip-net series. These and all other fry captured in excess of size/weight sample needs will be passed through a 2-in tunnel coded-wire tag detector (supplied by ADF&G). Tagged fry will be frozen individually in clean labeled scintillation vials for later pick-up by ADF&G. Fry retained for size and weight measurements will be suffocated (not preserved) prior to same-day processing at the hatchery. The fork length of each fry will be measured to the nearest mm and blotted fresh weight recorded to the nearest mg. Every eighth fish will be dissected for stomach contents (25 per 200 fry sample).

3. Fry stomach contents: Each fry selected for stomach analysis will be opened carefully and stomach contents removed for identification. Because fry tend to regurgitate food items if preserved in formalin, fish will be suffocated in a closed jar prior to analysis. Food items will be identified to the lowest taxon possible (generally species, sometimes genus, occasionally family or order) and counted. The mass of each food category will be estimated using the mean weight of representative species or composites obtained from the literature or determined in the laboratory from specimens in plankton tows (Coyle and Paul 1988, 1989). This practice avoids problems with weighing partially digested food items. Numbers, kinds and masses of food will be reported for each fry in a sample.

4. Small-scale fry migrations: The location and estimated size of fry schools in and near Sawmill Bay will be recorded on standard field charts each time the study team is sampling for fry and their food. Numbers and kinds of dead and moribund fishes will also be noted on these records. Fishes that cannot be readily identified will be frozen and stored for later taxonomy.

5. Surface water temperature: A laboratory grade mercury thermometer will be used to measure the temperature of the water at approximately 0.5 m depth below the surface. Temperature will be recorded to the nearest 0.1 deg C. If a back-ordered lowering thermistor thermometer is acquired during the sampling season, temperatures at 2 m intervals from the surface to 30 m will be obtained at the two zooplankton vertical-net stations in and near Sawmill Bay.

LOGISTICS

A field team of three experienced marine science graduate students and one volunteer will conduct studies at and near Sawmill Bay by establishing a base of operations at the AFK Hatchery. These studies will be supported by skiffs, food, lodging and laboratory space provided by the Prince William Sound Aquaculture Corporation. Studies will begin on May 8 and continue through June 29 when most fry are expected to have moved away from near-shore environments. The study leader will assist with the establishment of sampling protocol in the field and will establish standard laboratory procedures and verify plankton identifications.

A daily log of observations and analyses will be kept on site and generalized weekly reports of progress sent by FAX to the University of Alaska. Copies of weekly reports will also be filed with the hatchery manager and the PWSAC main office in Cordova.

Hand-held VHF FM marine radios will be purchased so that the study team can maintain contact with the hatchery (if necessary) during field operations. Life preservers for each person will be obtained from the Seward Marine Center.

DATA ANALYSIS

Salmon forage fields: The patchy nature of zooplankton populations, associated with physical features, predation pressure and growth, is addressed by replicating samples in time and space. The results from previous extensive studies of inshore plankton communities near Kodiak Island (Vogel and McMurray 1986) indicate that, for the species comprising the bulk of salmon fry food, approximately 5 samples are needed to detect differences of a factor of 2.0 (alpha = 0.05) at specific times and places. Duplicate samples are expected to detect differences of a factor of 3.0 or more. Five or more locations at non-oiled and oiled sides of Prince William Sound were selected for samples of open-water forage stocks. To conform with previous sampling procedures at locations near AFK, three or more sets of samples were planned each week. Replicate tows will provide measures of sampling error associated with collections from a given water column. Samples within regimes by cruise or week will provide

measures of variability needed to test hypotheses of no detectable difference between oiled and non-oiled sites.

One-way and complex Analysis of Variance models will be used to test hypotheses and to provide estimates of variability associated with sampling a water column, and sampling a specified time period (week) and location. This information will be vital in determining the number of samples needed to achieve specified levels of detecting differences (alpha=0.05) between treatment groups in possible continuing studies of oil impact.

To meet the assumptions of normality and independence of mean size and variance, plankton samples will be transformed by base 10 logarithms prior to ANOVA (Taylor 1953). This is standard practice in plankton ecology, yielding geometric means and variances needed for hypothesis testing and more efficient allocation of resources.

Fry food comparisons: Stomach data will be pooled for individual 25-fry samples and grouped by week and location (inside and outside Sawmill Bay). An index of Relative Importance (IRI; Pinkas et al. 1971) will be used to characterize fry diet preferences and to rank prey items. A Percent Similarity Index (PSI; Whittaker 1952) will be employed to determine diet overlap at oiled and non-oiled sites and to compare food preferences with historical data. The sampling design will generate approximately 750 fry stomach analyses over the course of the 10 week study.

Salmon fry growth: Estimates of fry growth as percent body weight per day will be obtained assuming exponential growth:

$$Ln\ (Wt) = gt + Ln\ (Wo)$$

where Wo and Wt are initial and later fry weights respectively over some interval of time, t days, and g is the instantaneous rate of daily growth. A linear regression fitting the natural logs of observed weights to a time base in days provides an estimate of g which when multiplied by 100 can be expressed as percent body weight gain per day (% bw/d). Estimates of this parameter (Mortensen and Wertheimer 1988) for unaffected fry populations in Auke Bay, Alaska range from 3.0 to 5.0 % bw/d.

Since fry at AFK enter Sawmill Bay from protective net pens in huge groups of several tens of million each at intervals of a few weeks, two or more distinct size-groups of fry may be in residence at any time and place. Length-frequency histograms will be examined to determine whether different modes of fry are present in samples. An attempt will be made to calculate the growth of distinct cohorts whenever possible, since lumping all fry will tend to negatively bias observed growth. Also, the larger fry in any population are presumably able to more effectively evade capture leading to under-representation in dip-

net and beach seine samples. This means that growth rates calculated from untagged lots of fry will suffer negative bias, particularly late in the season when the fry become larger and more active. This error is not expected to negate comparisons with historical and present data sets (ADF&G, NMFS) where % bw/d is calculated from untagged fry captured using similar methods.

SCHEDULE

The following tabulation describes the milestone dates and activities planned for the course of the study, April 1, 1989 through February 28, 1990:

DATE	ACTIVITY
5-11 April	Research cruise HX 121; Zooplankton sampling in Prince William Sound.
20 April	Joint ADF&G, NMFS and UAF Salmon fry study planning meeting in Cordova.
5-10 May	Research cruise HX 123; Zooplankton sampling in Prince William Sound.
8 May	Research team on site at AFK; sampling initiated.
12-14 May	Study Leader at AFK Hatchery to finalize sampling protocol.
1-7 June	Research cruise HX 125; Zooplankton sampling in Prince William Sound.
29 June	Research team departs AFK hatchery. All plankton and fry length, weight and stomach content samples processed.
5 July	All records of summer activities and data logs delivered to the principle investigator.
7 July	Meeting of the research team and study leader to discuss the summer's activities and to assign data analysis tasks.
16 August	Meeting of the research team to report progress on data analyses.
1 November	Preliminary data analysis complete, draft of data report and synthesis prepared.
15 November	Meet with ADF&G and NMFS personnel to develop SOPs and plan the 1990 field season.
1 January	Final draft of Prince William Sound and AFK studies report.
28 February	Proposal completed for 1990 study.

BUDGET/PERSONNEL

Personnel (100)	$94,100
Travel (200)	$7000
Services (300)	$29,700
Commodities (400)	$6500
Equipment (500)	$4000
Overhead (700)	$28,300
Total	$169,600

Personnel

The study leader is funded for 3.0 months. Three full-time graduate students (Ms. Ashley Evans, Ms. Elizabeth Stockmar and Mr. Loren Tuttle will share portions of the graduate student support. Four 3-month temporary positions will be recruited for field and/or laboratory assistance when needed.

R. Ted Cooney; study leader	3 man months
A. Evans	12 man months
E. Stockmar	6 man months
L. Tuttle	6 man months
TBF four temporary lab/field assistants	12 man months

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Figure 1. Study sites used in component study I, Impacts of oil spill on migratory behavior, growth and mortality. The five hatcheries are shown in bold letters. The numbers refer to the six sample collection sites.

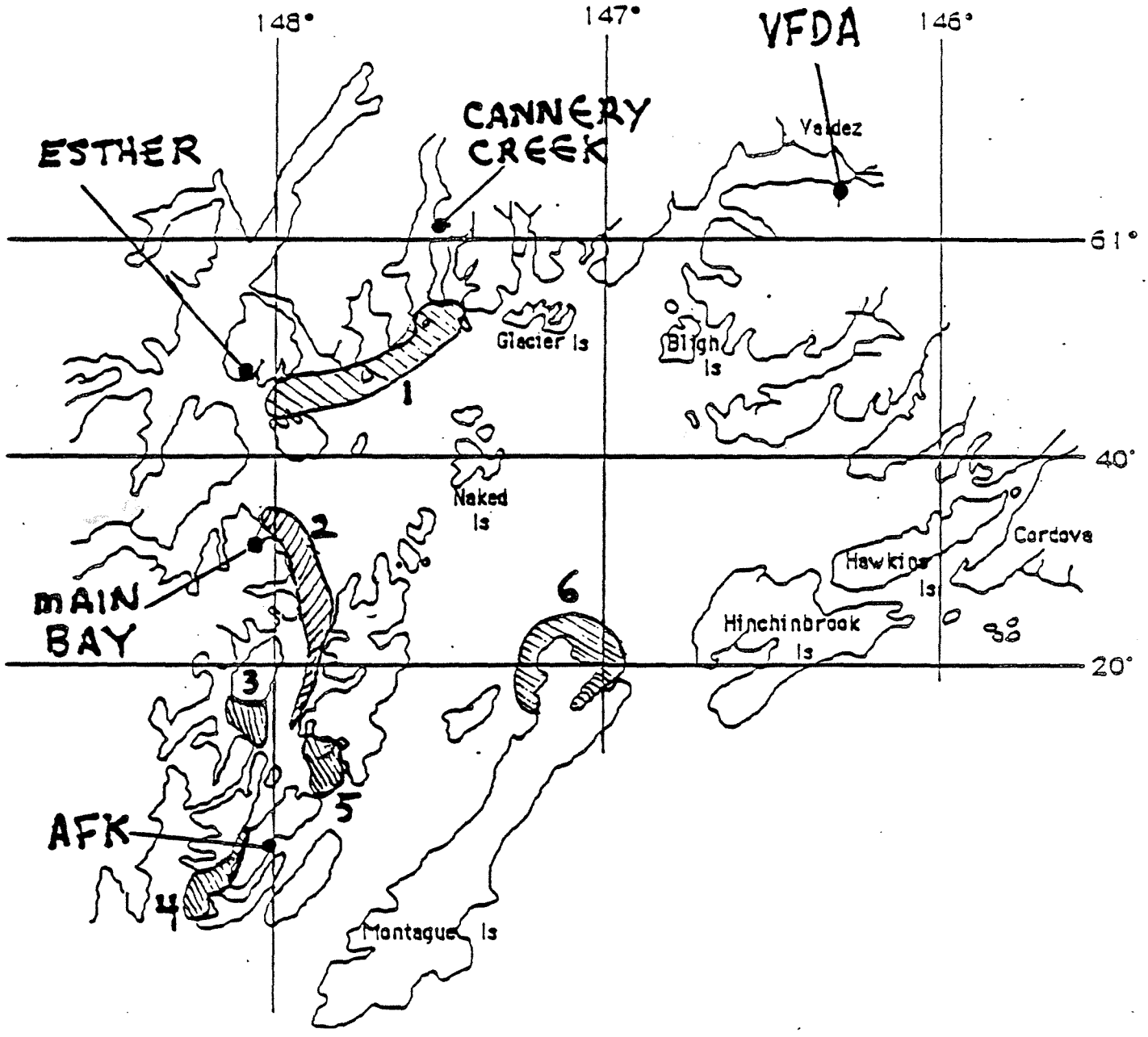
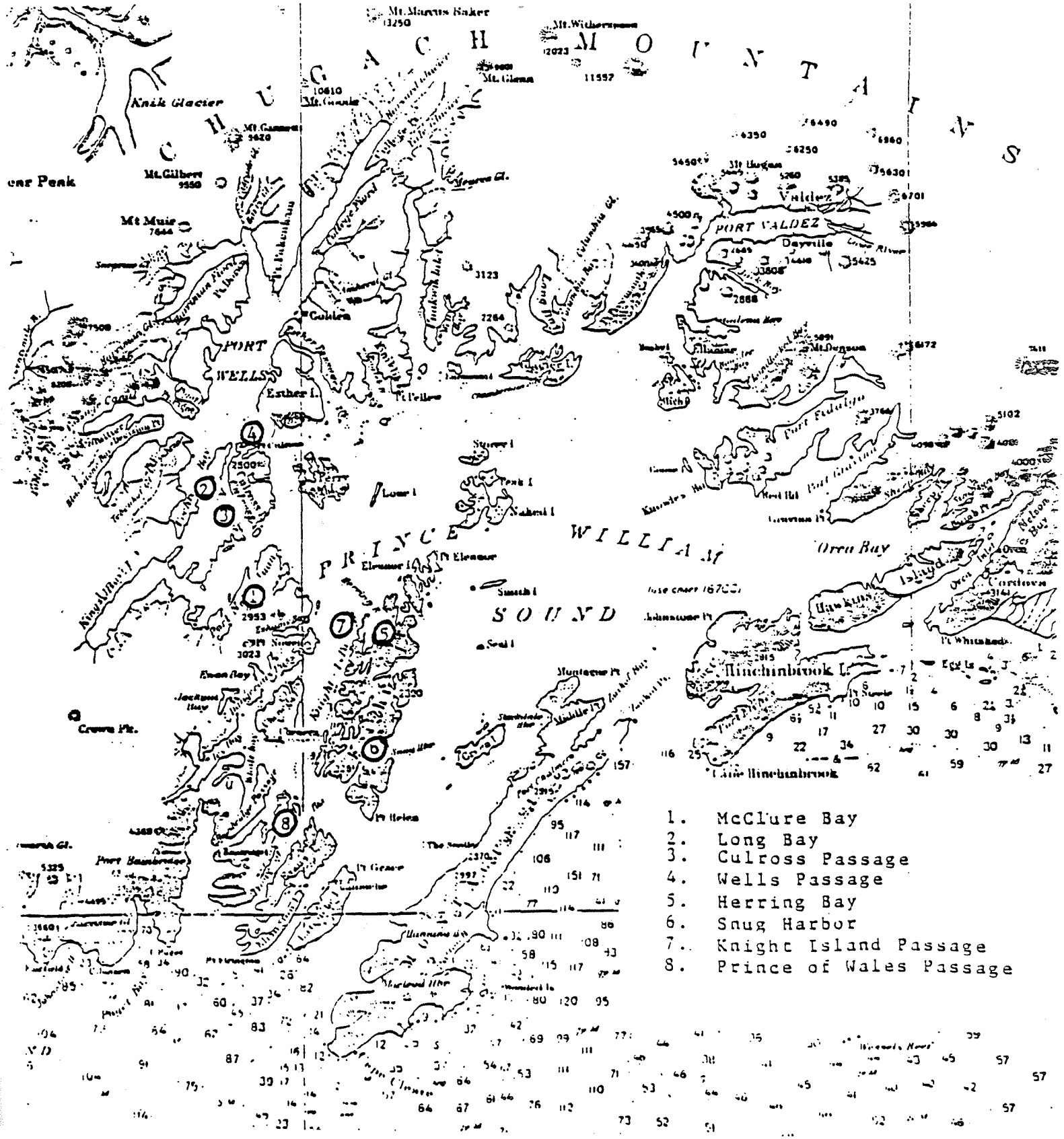


Figure 2.
Map of Prince William Sound showing location of study sites.

Component II: Impact of oil spill on juvenile pink and chum salmon and their prey in critical nearshore habitats.



STATE/FEDERAL RESOURCE DAMAGE ASSESSMENT
DETAILED STUDY PLAN

CONFIDENTIAL

DRAFT

Project Title:	INJURY TO DOLLY VARDEN CHAR AND CUTTHROAT TROUT IN PRINCE WILLIAM SOUND
Study ID Number:	Fish/Shellfish Study Number 5
Lead Agency:	State of Alaska, ADF&G; Sport Fish Division
Cooperating Agencies:	Federal: USFS State: DNR
Principal Investigator:	Kelly Hepler, Fishery Biologist
Assisting Personnel:	Andrew Hoffmann, Fishery Biologist Tom Brookover, Fishery Biologist Nine Fishery Technicians
Date Submitted:	September 25, 1989

	Signature	Date
Principal Investigator:	Kelly Hepler	10/9/89
Supervisor:	Doug Eggli	10/9/89
OSIAR Senior Biometrician:		
OSIAR Program Manager:	Richard Holmes	10-13-89
OSIAR Director:		

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INTRODUCTION

The goal of this study is to compare the abundance, survival, and growth of populations of cutthroat trout Oncorhyncus clarki (hereafter referred to as trout) and Dolly Varden char Salvelinus malma (hereafter referred to as char) differentially affected by the oil spill in Prince William Sound. Trout and char are estuarine anadromous species that inhabit PWS (Morrow 1980). Unlike anadromous Pacific salmon *Oncorhynchus spp.*, trout and char utilize near-shore and estuarine areas for feeding. Their marine migrations are not as extensive as those of Pacific salmon (Morrow 1980). Some of the most important stocks of these species inhabit areas that have been severely impacted by direct contact with oil including Green and Montague Islands and Eshamy Bay (Mills 1988). Since these species commonly live to age 8 (Morrow 1980), the potential exists for both short-term and long-term effects from exposure to oil. Study of these species is crucial in that they represent the only finfish species in the fishery assessment proposal package that inhabit the most oil-affected areas (the near-shore waters of the sound) throughout most of their lives.

Life History

The experimental design for this program is based upon the model developed by Armstrong (1970, 1974, 1984) and Armstrong and Morrow (1980) to explain the migratory behavior of anadromous char. This model identifies two patterns of life history: fish that were spawned in lake systems and fish that were spawned in non-lake systems. For both groups, juvenile char remain in freshwater residence in their natal stream for up to four years. During their last spring of freshwater residence, they smolt to sea. During late summer or early fall, fish that were spawned in lake systems return to their natal stream to overwinter in the freshwater lake. During the spring, they again emigrate into marine waters and annually return to their natal lake system during late summer or early fall to spawn and overwinter. Fish that were spawned in non-lake systems exhibit a more complex migration. Upon smolting, juvenile char search for a lake system to overwinter. These fish then behave in the same manner as do fish that originate in a lake system except that they return to their natal stream to spawn and then return to their selected lake system to overwinter.

The migratory habits of anadromous cutthroat trout are more poorly understood than those of anadromous char although it appears that they exhibit similar migratory habits to char (Jones 1982). Trout, however, spawn in the spring as opposed to fall for char.

Experimental Design

It is hypothesized that two detrimental impacts on these species could result from the presence of large amounts of crude oil in marine waters including: (1) reduced survival; and (2) reduced growth. To test

whether there will be a measurable impact on these stocks, two to four lake systems having stocks of trout and char from each of two treatments were selected for study. A high-impact treatment is defined as a stock emigrating from a lake system which flows into a marine environment which has been directly impacted by oil, while a low-impact treatment is defined as a stock exiting from a lake system into an area which has not been directly impacted by oil.

The principal objective of the project is to measure annual abundance, survival, recruitment, and growth of the stocks of char and trout in each of the study streams over three calendar years. Our primary assumption is that there is a difference in exposure to oil for fish stocks from each of the two treatments. Evidence from the literature indicates that marine migrations can range up to 116 kilometers for char (Armstrong 1974) and 80 kilometers for trout (Jones 1982). Although, marine migrations from low-impact waters may extend into oiled marine waters, the stocks selected within each treatment still represent different treatments in that the marine waters first encountered upon entry from freshwater will be very different in their oil content. Thus, it is assumed that any significant changes in stock abundance, composition, or dynamics from the initial emigration of stocks in the high-impact treatment as compared to stocks from the low-impact treatment is due to contact with the oiled marine waters.

Armstrong's model of migratory behavior provides the basic framework for this study. First, each of the study streams represents a stock of fish that annually homes to that specific overwintering stream. Second, since overwinter residency occurs entirely in freshwater, fish sampled during the spring emigration from each stream have not yet encountered oiled waters. Given this, the first sample from each stream (the first emigration) will provide the baseline data for stocks in each stream and treatment.

A measurable detrimental impact on these anadromous stocks of trout and char may result in a loss to the sport fishery. The status of the sport fishery will be investigated through (1) an ongoing postal survey (Mills 1988); and (2) an on-site creel survey of selected Prince William Sound fishery access ports (OSIAR Study FS #6).

OBJECTIVES

During 1989, the specific objectives of this project are:

1. to test the hypothesis that there is no difference in annual survival rates of char and trout between oiled and non-oiled lake systems (the test will be done given a level of significance of $\alpha = 0.05$.);

2. to test the hypothesis that there is no difference in annual growth rates of char and trout between oiled and non-oiled

lake systems (the test will be done given a level of significance of $\alpha = 0.05$.); and,

3. to test the hypothesis that fish from non-oiled systems do not migrate into oiled waters (the test will be done given a level of significance of $\alpha = 0.01$).

4. to assess exploitation rates in recreational fisheries of Dolly Varden char and cutthroat trout overwintering in oiled and non-oiled areas (through tagging in this project and recoveries from the creel survey project; specific methods are addressed in the creel survey project).

5. identify potential alternative methods and strategies for restoration of lost use, populations, or habitat where injury is identified (to be accomplished upon completion of this project).

To accomplish these objectives, the following tasks will be performed:

1. count the number of emigrating trout and char through weirs placed on three oiled and two non-oiled streams during the period May through the end of emigration;

2. estimate annual survival rates for each of the study stocks using abundance data collected at the weirs; and,

3. estimate mean length of emigrating trout and char from each study stream (the estimates will be ± 10 mm of their true values 90% of the time).

METHODS

This operational plan addresses both the long range study design of this project and work that has already been accomplished as part of the initial fishery impact assessment work. Throughout the remainder of this document, work that has already been initiated will be identified. Because of the immediacy of the initial fishery impact assessment work, some of the experimental design had not been fully developed and certain aspects of the sampling were not initiated. Changes in sampling design will be identified for initiation in the future.

Study Design

There are virtually no previous quantitative data for these species in PWS to draw upon as a baseline. However, since the trout and char were still in freshwater residence at the time of the spill, the opportunity exists to sample these fish during their 1989 emigration prior to any potential exposure to an oiled marine environment and use this as the

baseline for each system. Therefore, in addition to comparisons between the oiled and non-oiled treatment streams, comparisons are also possible between subsequent years' data from streams within each treatment and the 1989 baseline for each stream and treatment. Stated as an experimental design problem, the experiment has two treatments (oiled and non-oiled streams). Within each treatment, there are study streams with stocks of trout and char that have years as a factor of the design.

Each study stream consists of a freshwater lake-river system that: (1) is a tributary to marine waters that were either impacted by large quantities of oil or received virtually no oil and (2) contains stocks of anadromous trout and char. The specific systems are: Rocky Bay Lakes (Montague Island), Green Island Lake (Green Island), and Eshamy Lake (Eshamy Bay) in the oiled treatment; and Boswell Bay Lakes (Hinchinbrook Island) and Makaka Lakes (Hawkins Island) in the non-oiled treatment.

A weir has been installed on each study stream prior to the initiation of the 1989 spring emigration. A sockeye salmon smolt weir is already in place on Eshamy Lake outlet as part of the Salmon Coded-Wire Tag Studies project and sampling for char and trout will be conducted in conjunction with this project. In addition, a sockeye salmon smolt weir will be installed on Jackpot Lake (an oiled system) during 1990 as part of the same project and trout and char sampling will also be conducted in conjunction with this project.

All emigrating trout and char at each weir site are being counted and measured from tip-of-snout to fork-of-tail to the nearest millimeter. All trout and char greater than 200 mm in length are being tagged with numbered Floy FD-68 anchor tags and having their adipose fin removed to estimate tag loss.

Survival will be estimated for smolt, immature, and mature char. Initially, all char up to 200 mm will be considered smolt. All char between 200 mm and 250 mm will be considered to have been immature the year before. Blackett (1968) found the average length of mature char to be in the range 250 mm and larger and the smallest spawning char sampled in Kodiak in 1988 were 250 mm. Given this, char greater than 250 mm in length will be initially classified as being mature. At the conclusion of this year's sampling, we will analyze length frequency data to identify more precise classifications for smolt, immature, and mature fish.

Starting in 1990, char greater than 199 mm will be tagged with Floy FD-68 anchor tags and char under 200 mm and greater than 109 mm will be tagged with numbered Floy Fabric tags. If all fish can be censused and examined for tags in all years, survival can be estimated for each system. Annual survival will be estimated for three groups: smolts, immature, and mature char. The mortality rate of spawning char is known to be high, particularly for males (Armstrong 1974). Therefore, the rate of survival estimated for smolt and immature char will be used to test the hypothesis of equal survival between oiled and non-oiled systems.

The hypothesis of equal survival will be tested using a chi-square statistic. The goals set in Table 1 for the number of fish to mark were calculated under the assumption that all char will be censused and examined for tags. However, if unknown numbers of fish can be expected to be lost past the weir (due to such events as weir washout), it will not be possible to directly estimate survival from the numbers released and returned. Instead survival will be estimated using mark-recapture methods. Estimates of survival (Seber 1982) from a mark-recapture experiment with their 95% confidence intervals at three levels of abundance were examined to estimate the sample goals required to detect significant differences in survival (Appendix A). Information from Armstrong (1974) indicates that survival of smolt to spawning is between 10 to 15 percent. For example, with an abundance of 30,000 fish and expected survival of 10%, a minimum sample goal of 12,000 fish would need to be tagged (Table 1). The percent of the emigration that should be examined for tags is also presented in Table 2. However, this is a fall-back position, in that a minimum number must be achieved if all fish cannot be handled due to external, uncontrollable factors. At the tagging level of 12,000 fish, if all fish are examined during the second and third springs, the test for independence will be able to detect differences in survival as small as 1-2% given 95% confidence (Table 2).

Estimates of survival will be adjusted, if necessary, for differential fishing mortality from the sport fishery. Fishing mortality for the various tag lots will be estimated using the methods of Clark and Bernard (1987). Examination of sport-caught fish for tags will be accomplished through the port sampling program conducted as part of the Prince William Sound and Gulf of Alaska Sport Fishery Harvest and Effort Project (Collingsworth *et al.* in print). Estimates of sport harvest will be obtained from an ongoing postal survey (Mills 1988).

The hypothesis of equal growth will be tested by analysis of individual growth rate. Incremental growth for individuals will be computed from recaptured fish. The experiment will be conducted as an Analysis of Variance with stocks of char or trout serving as replicates within the oiled treatments. Years, and possibly initial length and/or seasonal growth, will serve as factor in the design.

The hypothesis that fish from non-oiled systems do not migrate into oiled waters will be tested using the geometric probability distribution. Thus the experiment consists of a series of trials that is concluded when the first success is observed. Success is defined as finding one or more tagged fish from a non-oiled area in an oiled area. The sample goal ($\rho = .01$) is to examine 460 fish at each location for the presence of tags.

Data Collection and Reduction

The data collection will be divided into two seasonal periods: spring and summer/fall. During the spring sampling, weirs are being used to count and sample the emigration of trout and char from study streams.

Weirs are installed approximately 0.5 km upstream from the saltwater terminus of the streams. The weirs are being operated by a two-person crew from early May to mid-July. The weirs utilize inclined screen panels using 5 cm poultry cloth screens and an overlay of 1.25 cm plastic mesh. The panel frames are constructed from wood and will be 2.0 meters long and 1.0 meters high. The panels rest against wooden tripods spaced approximately two meters apart. Both upstream and downstream live traps are installed. All species captured in the downstream or upstream traps are being identified, counted, and the tag number of any tagged fish recorded. Date, sex (if identifiable from external maturation characteristics), and length (tip-of-snout to fork-of-tail to the nearest mm) are being recorded.

Additional sampling will occur in the summer and fall at selected estuarine locations in the oiled and non-oiled regions of PWS. The objective of this sampling is to determine presence/absence of fish from the non-oiled stratum in oiled waters. Oiled estuaries to be sampled include those in Port Chalmers, Jackpot Bay, Chenega, Culross Passage and Island, Port Nellie Juan, and Bay of Isles. Fish will be caught for tagging and biological sampling as described above using some combination of traps, beach seines, variable mesh gill nets, backpack shockers, and hook and line. At each sampling site, catches will be examined for recaptures from the study streams. During subsequent years of weir sampling, catches will be examined for recaptures from these additional sampling sites.

All data will be recorded on standard Division of Sport Fish mark-sense forms. All completed forms are visually scanned for errors and corrected as necessary. Corrected forms are sent to Anchorage for processing by optical scanning. Copies of the resultant data files and summary printouts are checked for errors and corrected as necessary. Data associated with uncorrectable errors will be deleted form the data file. Corrected data files are returned to Anchorage for archiving.

Data Analysis

Estimates of annual survival will be computed through analysis of tag returns. If all emigrating fish can be examined for marks, the estimates of annual survival (S) can be simply computed as:

$S = m_2/R_1$

where:

m_2 = number of fish recovered in year y+1
R_1 = number of fish tagged in year y.

The Jolly-Seber three-sample method (Seber 1982) will be used in the event that each emigrating fish cannot be examined at the weirs. Buckland's program RECAP (1980) will be used to generate the estimates and variances. The assumptions of the Jolly-Seber model are:

1. every fish, tagged and untagged, has equal probability of being caught;

2. every tagged fish has equal probability of survival to the next sampling event;

3. every fish sampled has equal probability of being returned to the population;

4. there is no tag loss; and,

5. all samples are instantaneous.

The sampling event for the purposes of the mark-recapture experiment is the emigration of trout and char past the weirs. All emigrating fish must cross the weir and therefore are assumed to be equally vulnerable to being sampled. The assumption of equal survival of tagged fish will be tested for the different tag groups and for the different length classes using chi-square statistics. Tag loss will be estimated for fish tagged in 1989, as all tagged fish will also be adipose fin-clipped. The last assumption is technically violated as the sampling event lasts through the entire migration. However, as each fish will only pass the weir once, and can only be sampled once, the emigration can be treated as an instantaneous sample of the whole population.

The hypothesis of equal survival will be tested using a chi-square test for independence. If the contingency table cannot be built; that is, if the char are not sampled at the weir during emigration during the second and third year of the experiment; the 95% confidence intervals of the survival estimated from the multi-year mark-recapture experiments will be compared to test for significant differences. In order to examine the effect of initial length on subsequent survival, the tests and

estimates will be stratified by tagging length, and if possible a logistic regression will be used to estimate this effect. The hypothesis of equal growth will be tested using Analysis of Variance. The hypotheses that fish from non-oiled systems do not migrate to oiled systems and vise versa will be tested by examining for the presence or absence of tagged fish from weir sites in each treatment in samples from weir sites in the other treatment.

Annual individual growth will be calculated from the tag data as the difference between length at time of release and length at time of recovery. An Analysis of Variance will be used to test for significant differences in growth between fish from oiled and non-oiled areas. Variation due to differences in years and initial length can be controlled for through the use of a block and covariate in the linear model if necessary.

The assumptions of Analysis of Variance are:

1. random sample,
2. normal distribution, and
3. homogeneity of variance.

The assumption of normality will be tested using Kolomogorov's D statistic. In all likelihood the data will not be normally distributed and a logarithmic or a rank transformation will be necessary.

The homogeneity of variance assumption will be tested with a Barlett's test. Again, if the assumption is not valid a transformation will be used.

SCHEDULES

A schedule of tasks to be completed during 1989 is as follows:

Task	Dates
Weir Operation	4/20-7/15
Summer and Fall Sampling	5/01-9/15
Data Analysis and Report Preparation	9/15-2/15

REPORTS

Results of these study efforts will be reported to the Division of Oil Spill Impact Assessment and Restoration. Upon completion of litigation, these data will be published as either an Alaska Department of Fish and Game, Sport Fish Division, Fishery Data Series report or in the fisheries literature.

BUDGET SUMMARY

A line item breakdown of project costs beginning April 1, 1989, and ending February 28, 1989 are as follows:

Line Item	Category	Cost(thousands)
100	Personnel	218.1
200	Travel	7.0
300	Services	65.0
400	Commodities	79.4
500	Equipment	67.9
	Total	437.4

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Table 1. Number of Dolly Varden char that need to be marked and the minimum needed to be examined for marks in order to estimate survival in a 3-sample Jolly-Seber experiment (refer to Appendix A).

Mean Abundance	Survival	Minimum Number of New Tags to Deploy	Minimum Number of Fish to Examine	95% C.I. (percent)	Relative Precision
10,000	5%	8,500	75%	0.036-0.064	57
	10%	8,500	75%	0.071-0.129	29
	20%	8,500	75%	0.171-0.229	15
	50%	8,500	75%	0.471-0.529	6
30,000	5%	12,000	50%	0.030-0.066	33
	10%	12,000	50%	0.080-0.102	17
	20%	12,000	50%	0.170-0.221	10
	50%	12,000	50%	0.379-0.521	4
50,000	5%	15,000	50%	0.032-0.068	36
	10%	15,000	50%	0.082-0.118	18
	20%	15,000	50%	0.181-0.219	9
	50%	15,000	50%	0.467-0.533	7

Table 2. Number of Dolly Varden char smolts that need to be marked to test the hypothesis of equal survival, assuming spring emigration is 30,000 smolt. The assumption is made that all emigrants can be examined for tags in order to classify mortality versus survival.

From a Survival of	Given an alpha level of	To detect a difference of: 5%	10%	20%	25%	50%
15%	0.01	30,000	10,500	3,000	1,200	300
	0.05	19,500	6,000	1,500	900	300
	0.10	10,500	3,000	750	750	300
10%	0.01		10,500	2,100	1,500	300
	0.05	19,500	6,000	1,500	900	300
	0.10	10,500	3,600	900	600	300
5%	0.01		18,000	4,500	3,000	600
	0.05		10,500	3,000	1,500	300
	0.10	30,000	7,500	1,800	1,200	300

Appendices

Appendix A.

Calculation of sample sizes for estimating survival.

Appendix A. Calculation of sample sizes for estimating survival.

A major objective of the study is to estimate survival and test the hypothesis that there is no significant difference between oiled and non-oiled areas. If all fish can be examined at all weirs in the second year of the experiment, then this hypothesis will be tested using a contingency table and a chi-square statistic. Sample sizes for marks needed to achieve a test of size 0.05 were estimated by running the analysis based on different survivals and changes in survival (Table 1). In order to be able to perform this type of test, the goal of the projects will be to examine all fish emigrating, while the number tagged will be determined as described below.

If fish are able to pass the weir unaccounted for, e.g. during a flood, then the above method cannot be used, and we will need to estimate survival using a multi-year mark-recapture method. A three-sample Jolly-Seber experiment will provide an estimate of survival from the first to the second year, θ_1 (Seber 1982). The equation for estimating this survival is as follows:

$$\theta_1 = \left\{ \frac{R_2 \, z_2}{r_2} + m_2 \right\} / R_1$$

and its variance is:

$$\mathrm{Var}(\theta_1) = \theta_1^2 \left\{ \frac{(M_2-m_2)(M_2-m_2+R_2)}{M_2^2} \left\{ \frac{1}{r_2} - \frac{1}{R_2} \right\} + \frac{1-\theta_1}{M_2} \right\}$$

where:

R_i = the number of tags (new and old) released in year i.

r_2 = the total recoveries from release R_2 in future years, i.e. here in year 3,

z_2 = recoveries from char tagged in first year, not recovered in year 2, but recovered in year 3,

M_2 = number of tags present prior to the second years sampling, i.e. survivors of the first years release.

m_2 = number of tags recovered in second years sample.

and:

$R_i = N_i \, p_c \, p_t$

$M_2 = R_1 \, \theta_1$

$m_2 = M_2 \, p_c$

$$r_2 = R_2\ \theta_1\ p_c$$

$$z_2 = R_1\ \theta_1\ (1-p_c)\ \theta_2\ p_c$$

where:

N_i = abundance in year i

p_c = probability of capture in each years sampling, which is assumed in this analysis to be equal for all three years

p_t = proportion of fish captured that are tagged, also assumed equal for all samples

θ_2 = survival from year 2 to 3, which is assumed to be equal to θ_1.

The hypothesis that survivals are different between oiled and nonoiled areas at an alpha level of 0.05, can be tested by examining the 95% confidence intervals, if they do not overlap then the difference is significant. This exercise involved setting an expected level of abundance and survival, examining the confidence intervals achieved at different levels of tagging ($R_i = N_i\ p_c\ p_t$) and handling ($n_i = N_i\ p_c$) and choosing the sampling levels that allow us to detect a pre-defined difference in survival rates. This difference was set at 5%, and the results for three different population levels, and four levels of survival are presented in Table 2.
The tagging levels were increased by 10% to allow for sampling error in the tag recovery. The variance, and the confidence interval, depends to a great extent on m_2, r_2 and z_2, the tag recoveries in the second and third year (see equation 2). The percent of the emigration to be examined for tags that is presented in Table 2 is a minimum. The goal will be 100%, however the realized percent should not be allowed to fall below those presented in Table 2, if the objective as stated above is to be achieved.

STATE/FEDERAL RESOURCE DAMAGE ASSESSMENT
DETAILED STUDY PLAN

CONFIDENTIAL

Project Title:	PRINCE WILLIAM SOUND AND GULF OF ALASKA SPORT FISHERY HARVEST AND EFFORT
Study ID Number:	Fish/Shellfish Study Number 6
Lead Agency:	State of Alaska, ADF&G Sport Fish Division
Cooperating Agencies:	Federal: USFS State: DNR
Principal Investigator:	Craig Whitmore, Fishery Biologist III
Assisting Personnel:	Kent Roth, Fishery Biologist II Wilson Potterville, Fishery Biologist II John B. Murray, Fishery Biologist III 12 Fishery Technicians
Date Submitted:	September 25, 1989

DRAFT

APPROVED

Titles	Signature	Date
Project Biologist	Kent Roth	10/10/89
Supervisor:	Craig Whitmore	10/13/89
OSIAR Senior Biometrician:		
OSIAR Program Manager:	Robert P. Holmes	10/13/89
OSIAR Director:		

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INTRODUCTION

The recreational fishing industry is a vitally important component of the local economies of Valdez, Whittier, and Homer. Recreational fishing opportunities are also important to residents of and visitors to Cordova and Kodiak. During 1987, Prince William Sound (PWS) supported approximately 81,200 angler-days of recreational fishing which was approximately 65% above the 1980 to 1984 average of 47,000 angler-days (Mills 1988, 1981b-1985). The majority of this effort was concentrated in and adjacent to the communities of Valdez, Whittier, and Cordova, but an increasing number of recreational fishermen have been traveling to the remote waters of PWS.

On 24 March, 1989 the oil tanker Exxon Valdez ran aground on Bligh Reef, spilling approximately 11 million gallons of North Slope crude oil into the waters of PWS. In the following two months, the oil spread out of PWS and washed up on beaches of Cook Inlet and around Kodiak and Afognak islands. Prior to the oil spill PWS, Homer, and Kodiak sport fisheries were expected to continue expanding in both effort and harvest. A concurrent increase in the number of charter boats catering to sport fishermen was also anticipated. Loss of fish abundance, major shifts in fish distribution, loss of the pristine character of the area, and other associated impacts due to the spill of oil could result in a substantial decrease in participation in the recreational fisheries of PWS, and may also affect sport fisheries in Homer and Kodiak. This could lead to a serious loss of revenue to the local communities.

In order to determine if recreational fishermen have been impacted by the oil spill or by the publicity associated with the oil spill, anglers will be surveyed at major access points to the PWS, Homer, and Kodiak areas, and at Eshamy Lagoon in western PWS. Anglers will be asked where they fished and how many fish they caught, and their addresses will be taken if they are willing to respond to a follow-up mail questionnaire. The number of tagged and untagged Dolly Varden char *Salvelinus malma* and cutthroat trout *Oncorhynchus clarki* will be noted in the sampled harvest (information to be used by Project Nos. five and ten). Anglers at Eyak River, near Cordova, and at two sites on the Copper River Delta will also be interviewed to determine the tagged to untagged ratio of Dolly Varden char and cutthroat trout in the sport harvest.

Anglers access the sport fishing waters of PWS by road, boat, float equipped aircraft, and train. A substantial number of the anglers also use the service of charter boat operators or air taxi operators. Species sought by recreational fishermen include all five species of Pacific salmon *Oncorhynchus spps.*, Pacific halibut *Hippoglossus stenolepis*, rockfish *Sebastes* and *Sebastolobus spps.*, Dolly Varden char, and cutthroat trout. Wild stock and hatchery produced salmon contribute to the harvest in all major fisheries.

The recent spill of oil in Prince William Sound may impact the groundfish stocks in the PWS, Homer, and Kodiak areas. Groundfish harvested by sport anglers include rockfish, lingcod *Ophiodon elongatus*, and Pacific halibut. Rockfish stocks are semi-pelagic and associated with reefs and pinnacles. Lingcod are benthic, sedentary, and associated with reefs. Pacific halibut are benthic and migratory. Oil contamination of benthic environments could

kill these fish or chronically taint them due to persistence of oil in their environment or their food web. The presence of any oiled fish may cause a drop in fishing effort due to perceptions of unpalatable fish or may cause a drop in harvest due to both lethal and sub-lethal effects of ingested oil on fish. Therefore, a major part of this project will be to determine the species composition of the groundfish harvest in the marine sport fisheries of PWS, Homer, and Kodiak; and the incidence of oil contamination in these harvests.

A description of the major fisheries of PWS and concerns regarding these fisheries follows.

Valdez

The community of Valdez, located on the north shore of Port Valdez, provides access to Valdez Arm and the waters of northern PWS. Valdez is accessed by road and is home port for privately owned pleasure boats and a growing charter fleet. The community of Valdez has a population of 5,000 and is the exit port of the Alyeska Pipeline Terminal.

Valdez Arm supports the largest sport fishery in PWS and the largest pink salmon *O. gorbuscha* sport fishery in Alaska. Other fisheries of importance include those targeting coho salmon *O. kisutch*, Pacific halibut, Dolly Varden char, and numerous species of rockfish. Chinook salmon *O. tshawytscha* have been stocked in Port Valdez since 1985 but have yet to create a substantial sport fishery.

Valdez Arm supported an average of 17,142 angler-days of sport fishing effort annually during the years 1980 through 1984 (Mills 1981b-1985). This effort increased to an average of 37,270 angler-days for 1985 and 1986, and to 45,745 angler days during 1987 (Mills 1986-1988). Sport fishing effort during 1989 is expected to be similar to the 1987 level. The increase in angler-days during 1987 and 1988 can be attributed to successes in pink salmon production by the Valdez Fisheries Development Association's (VFDA) Solomon Gulch Hatchery, a private non-profit facility (Roth and Delaney In press).

In addition to the sport fishery for pink salmon, Valdez Arm supports the largest and most consistent sport fishery for coho salmon *O. kisutch* in PWS. Sport anglers harvested an average of 6,500 coho salmon annually during the years 1983 through 1987 (Mills 1984-1988). The Valdez Arm coho salmon fishery is supported by both natural and VFDA hatchery-produced coho salmon. Valdez Arm also supports the largest Pacific halibut fishery in PWS. The estimated sport harvest of Pacific halibut in Valdez Arm has increased from 339 in 1978 to 3,241 in 1986 (Mills 1980, 1987). The 1986 harvest of Pacific halibut in Valdez Arm accounted for 39 percent of the PWS harvest for that year (Mills 1987).

Whittier

The community of Whittier provides access to Passage Canal and the waters of western PWS. Whittier is accessed primarily by railroad from Portage or float plane from Anchorage. In recent years, it has become the home port for many privately owned pleasure boats and a growing charter fleet. Sport effort in the Whittier/Passage Canal area has increased over 100% from 1977 (Mills 1979, 1988).

Western PWS produces numerous strong runs of pink salmon and chum salmon *O. keta*. No native chinook salmon are present, coho salmon are scarce, and sockeye salmon *O. nerka* are only present in select lake systems. In an effort to increase sport fishing opportunities in western PWS, coho salmon smolt have been stocked in Passage Canal annually since 1978, and chinook salmon smolt stocking began in 1981. The stocking programs have provided good angling opportunity in the Whittier Terminal area of Passage Canal. The coho salmon stocking program was expanded to include Culross Lake in 1983 and Surprise Cove Lakes in 1985. In 1986, juvenile chinook salmon were stocked into two Granite Bay lakes (Esther Island).

Cordova

The community of Cordova provides access to the waters of eastern PWS. Cordova is primarily a commercial fishing community. Recreational fishing from this community is primarily conducted along the road system near Clear Creek, downstream of Eyak Lake, and from the beach near Flemming Spit. In 1989, freshwater drainages crossed by the Copper River highway were opened to sport fishing for salmon. Fishing effort is directed toward sockeye salmon, pink salmon, cutthroat trout, Dolly Varden char, Pacific halibut, and rockfish. Additionally, boats and float equipped aircraft travel from Cordova to various locations in eastern PWS in pursuit of recreational fishing activities.

Harvest and effort data for these fisheries are limited to information gained from the state-wide mail survey. These data indicate that both sport effort and harvest are increasing annually (Mills 1979-1988).

Eshamy

The Eshamy system supports one of the most important sockeye salmon stocks of western PWS. The lagoon and lake areas of the Eshamy system are one of the few locations where sport fishermen can harvest sockeye salmon in western PWS. Sport fishermen access the area either by boat out of Whittier or by float plane from Anchorage or the Kenai Peninsula. Oil from the Exxon Valdez has spread into the Eshamy area.

Seward

The recreational fishery in Resurrection Bay is one of the largest marine sport fisheries in Alaska (Mills 1988). Most of the effort in the fishery is by private boat anglers, however there is also a large charter boat fleet. Most of the sport fishing effort is directed toward coho salmon, however Pacific halibut, rockfish, lingcod, and chinook salmon are also targeted. Shore anglers fish for coho salmon, chinook salmon, and a few Dolly Varden char. The coho salmon and chinook salmon populations are supplemented by an extensive stocking program.

Weather and water currents saved Resurrection Bay itself from heavy oil contamination, however isolated patches of oil washed ashore in Resurrection Bay on beaches as far north as Lowell Point near Seward. Areas frequented by anglers fishing for rockfish and halibut outside of Resurrection Bay were exposed to heavy contamination.

Homer

Sport fishing and tourism are major components of the Homer area economy. Kachemak Bay and nearby waters support the largest Pacific halibut recreational harvests in Alaska. In 1987, charter boats harvested an estimated 20,319 Pacific halibut in the Kachemak Bay area, and private boats harvested an additional 21,369 (Mills 1988). In 1989, it is anticipated that there will be over 100 charter boats fishing for Pacific halibut out of Homer, along with a large fleet of private boats. The marine waters of Kachemak Bay also support major fisheries for stocked chinook salmon in Halibut Cove; for stocked pink salmon in Tutka Bay; and for stocked chinook salmon, coho salmon, and pink salmon near the Homer Spit. Oil from the Exxon Valdez reached the Kachemak Bay area nearly two months after the spill. Its impact in the area has been spotty, with weathered oil washing up in isolated locations. Many halibut boats from Homer, however, fish past Seldovia towards the outer waters of the Kenai Peninsula which were more heavily hit by oil.

Kodiak

The city of Kodiak is a major commercial fishing port. The city also supports a large US Coast Guard base. Sport fishing is a relatively minor component of the local economy, but is an important recreational activity for local residents and Coast Guard personnel. There are estimated to be fewer than 15 charter boats operating out of the Kodiak harbors. Sport fishermen from the Kodiak area harvest mainly Pacific halibut, pink salmon, coho salmon, and rockfish in marine waters. Anglers fished an estimated 38,671 days in marine waters of Kodiak in 1987 (Mills 1988). The eastern side of Kodiak Island was not hit by the oil until over two months after the spill, however storms at that time spread the oil in isolated locations all around Kodiak Island. Commercial salmon fisheries in the Kodiak area have been delayed or curtailed by the presence of oil in fishing areas, and these impacts have been given much attention in the press.

Anchorage

Anchorage has the largest float plane base in Alaska and is the operation center of approximately 30 charter service companies that frequently provide transportation to PWS for recreational fishermen. Information regarding the extent of these operations, the areas and stocks they target, or the effort and harvest rates is currently unknown. However, it is believed that these operations may constitute a significant component of the sport fishery of PWS.

OBJECTIVES

During 1989, this program will be operational during the period late June to mid September. Specific objectives of the FY90 investigations are:

1. Estimate recreational catch and harvest of salmon, rockfish, halibut, cutthroat trout, and Dolly Varden char. Specifically we will:

    a. Estimate the species composition of the rockfish harvest in the PWS, Seward, Homer, and Kodiak marine boat sport fisheries. Objective criteria are such that the estimated proportional contribution will be within ± 5% of the true proportion 95% of the time.

    b. Estimate catch and harvest per angler-day by species for sport anglers returning to major marine access points in PWS, Homer, and Kodiak. Objective criteria are such that the estimated catch and harvest per angler day are within ± 10% of their true values 90% of the time.

    c. Estimate the number of fish caught and harvested, by species, by sport anglers at Eyak Lake and two Copper River Delta streams during the period 15 June through 1 October 1989 such that the estimated catch and harvest are within ± 10% of their true values 95% of the time.

    d. Estimate the number of fish caught and harvested, by species, by boat and shore sport anglers in Eshamy Lagoon during the period 1 July through 4 September 1989 such that the estimated catch and harvest are within ± 7.5% of their true values 95% of the time.

2. Estimate angler effort and identify the temporal and spatial distribution and location of origin of angling effort. Specifically we will:

    a. To estimate sport effort (in number of angler-hours) by anglers at Eyak Lake and two Copper River Delta streams during the period 15 June through 1 October 1989 such that the estimated effort is within ± 10% of its true value 95% of the time.

    b. To estimate effort (in number of angler-hours) by shore and boat anglers in Eshamy Lagoon during the period 1 July through

4 September 1989 such that the estimated effort is within ± 7.5% of its true value 95% of the time.

3. To inspect enough groundfish and salmon such that there will be a 95% chance of finding at least one contaminated animal when at least one fish in 500 (0.005) is tainted.

4. Identify potential alternative methods and strategies for restoration of lost use, populations, or habitat where injury is identified.[1]

In addition to the objectives noted above, the following tasks will be accomplished:

1. To collect names and addresses of all interviewed anglers who are willing to respond to a follow-up mail questionnaire.

2. A logbook survey of sport anglers who access PWS from the float-plane base in Anchorage will be conducted to collect data on selected demographic characteristics and general catch and effort parameters.

3. To monitor sport harvests in PWS and Homer for Dolly Varden char and cutthroat trout tagged during population studies in PWS, Kodiak, and the Kenai Peninsula.

4. To collect age and length data from rockfish (by species), lingcod, and Pacific halibut harvested in the PWS, Homer and Kodiak marine boat sport fisheries.

PROCEDURES

Marine Catch Sampling

Study Design:

Marine catch sampling will be done by one fisheries technician at each of the following locations: Valdez boat harbor, Cordova boat harbor, Whittier, Seward, Homer, and Kodiak. The technicians will accompany the technicians responsible for the marine angler surveys, and will sample as many fish as possible. At Cordova, the same technician will also be entirely responsible for the marine angler survey, and at all other locations the technicians will assist with the marine angler survey as time allows.

Five hundred harvested rockfish will be identified to speciesduring each month of the marine sport boat fishery. This sample size should allow for the estimation of the species composition given the objective criteria stated in Objective 1 (Thompson 1987). Collection will be done by month to account for possible changes in the species composition of the harvest over the time the fishery operates.

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1 This objective will be accomplished at the completion of the project.

Over the course of the survey a total of 600 harvested groundfish (rockfish, lingcod, and Pacific halibut) and 600 harvested salmon will be examined for oil contamination. This sample size should result in a 95% probability of observing one contaminated fish assuming that the overall incidence of contamination is 1 in 200 (i.e., 0.005). This sample size was obtained by assuming that the incidence of contamination is distributed as a binomial random variable. Accordingly, the probability of observing at least one animal with the selected characteristic is given by:

$$1 - \text{Probability} = (1-p)^n \qquad [1]$$

where p = the rate of incidence of the characteristic in the population (assumed to be 0.005 in this case); and n is the sample size. Since we want this probability to be equal to 95%, then we solved for n as follows:

$$n = \log(1-.95) \div \log(1-.005) \qquad [2]$$

Which equals 597.65 or approximately 600 fish.

All examined fish will be noted as to their species and area of harvest.

One hundred fifty groundfish of each species examined for oil will be sampled for age and length. Groundfish to be sampled include all species of rockfish observed during the creel survey, lingcod, and Pacific halibut.

Data Collection:

Rockfish, lingcod, and Pacific halibut harvested by charter and private boat anglers will be examined for the presence of oil and will be measured for length and sampled for age. This sampling will be done in conjunction with the marine angler surveys.

In Homer and Kodiak bins will also be placed near fish cleaning stations for private anglers or charter boat employees to discard the carcasses of fish that are caught. Rockfish, lingcod, and Pacific halibut are commonly filleted with the remaining carcass (including head, gill, intestines) left intact. Lengths, otoliths, and gut samples can be taken from these carcasses. In Kodiak the bins will be adjacent to the Lions Club cleaning table at the small boat harbor and at the fish cleaning shed on the Coast Guard base. In Homer the bins will be placed near the fish cleaning tables of selected charter boat operations.

The total number of fish inspected by day and species will be recorded in a field notebook. Fish will be inspected for oil contamination as follows. The gills and body of the fish will be inspected for obvious signs of oil. Crude oil contamination can occur between scales or on descaled areas. The gills will further be inspected for odor and visual signs of crude oil. The gills are the most likely to be contaminated as they are slow to cleanse, even if the fish had moved into uncontaminated waters. The best indicator is visual identification. Severely contaminated gills should be dark brown in color.

The gills suspected to be contaminated will be wiped with a white paper towel. If present, oil should be discernable as small brownish particles on the paper towel. Care will be taken to not confuse contamination from crude oil with that of diesel oil. If present, diesel oil exhibits a much stronger smell than would crude oil at this stage of aging. Also, gill discoloration can be caused by decomposition.

The stomachs of rockfish, lingcod, and Pacific halibut will also be inspected for ingested oil (tarballs). If present, they may be extremely difficult to detect in that the tarballs may appear as small "B-B"-sized particles. It is doubtful that tarballs would exhibit a detectable odor.

Any fish or sample that is suspected of having oil contamination will be collected for further inspection. If possible, the entire fish will be collected. The sample will be wrapped in aluminum foil, marked with a locking evidence tag, logged, and stored in a locking freezer. The Alaska Department of Environmental Conservation will be contacted for further analysis. "Chain of Custody" procedures will be followed in the processing of these samples (see Appendix D).

Species identification of rockfish will be accomplished using the methods of Kramer and O'Connell (1988). Technicians will keep a daily tally of the number of each rockfish species observed and of the total number of rockfish observed.

Length measurements will be for total fish length to the nearest millimeter. Lengths will be recorded on AWL mark sense forms. Rockfish will all be recorded on the same form, using the "weight" field to differentiate species as described in Appendix B. Age structures to be collected will be otoliths for Pacific halibut and rockfish, and finrays for lingcod. The litho-code and line number for each fish from the mark sense form will be recorded on the envelope containing the age structure so that the length and age for each fish can be matched.

In Cordova, Valdez, Whittier, Eshamy, Homer, and Kodiak the technicians involved in the angler surveys and the marine catch sampling will watch for Dolly Varden char and cutthroat trout that have a missing adipose fin or are a marked with a floy tag. These fish were marked as part of damage assessment studies in PWS, Homer, and Kodiak (OSIAR Project Nos. five and ten). The technicians will keep a tally of the number of Dolly Varden char and cutthroat trout they have observed each day. Any marked fish that are seen will be measured and the tag number, tag color, length of the fish and area harvested will be recorded on a tag recapture form (Appendix B). Area harvested will be recorded with as much detail as possible.

Data Reduction and Analysis:

Mark sense forms will be visually examined for errors by the technicians and then sent to the Division of Sport Fish's Research and Technical Services group (RTS). RTS will optically scan the mark sense forms to produce a computer data file, a frequency report, and a listing of each batch of data.

The file and reports will be returned to the technician for editing and analysis. The final, edited data file will be returned to RTS for permanent storage on tape.

A contingency table analysis (Sokal and Rohlf 1981, Box 17.8, pages 745-746) will be used to test for differences in rockfish species composition over time. If none are found ($\alpha = 0.05$), the data will be grouped to estimate the species composition for the season. The proportion of fish of each species will be estimated along with its standard error using the procedures outlined by Cochran (1977, section 3.2, pages 50-53). Numbers of rockfish harvested by species will be estimated based on the estimated proportional contribution of sampled fish and the estimated sport harvest of rockfish from the Statewide Harvest Survey. The variance of these estimates will be estimated using Goodman's (1960) formula for the exact variance of the product of two independent random variables.

Age and length frequency for each species will be summarized. Proportions of each age class will be estimated using the procedures outlined by Cochran (1977, section 3.2, pages 50-53). Mean length at age with the associated standard errors will be estimated using standard statistical procedures (Sokal and Rohlf 1981, Boxes 4.2 and 7.1, pages 56 and 139).

Marine Angler Surveys

Catch and harvest per angler day, and selected characteristics of anglers participating in the marine boat sport fisheries of PWS, Homer, and Kodiak will be estimated using a stratified two-stage survey. A technician will be stationed at Valdez, Whittier, Homer, and Kodiak to conduct a survey of anglers returning to harbors at each location. In Seward, an on-going creel survey (Appendix E) will be used to collect information for this project. In that survey both boat and shore anglers are interviewed. The marine angler survey at the boat harbor in Cordova will be conducted by the technician who will also be responsible for marine catch sampling. The technicians will interview anglers for approximately 6 hours each day, working all weekend/holiday days and 3 weekdays each week.

Study Design and Data Collection:

Valdez. The survey of the marine sport boat fishery operating out of Valdez Harbor will be conducted from 15 June through 15 September 1989.

Selected angler characteristics,will be estimated using a stratified two-stage survey. At Valdez, the fishing day is defined as 14 hours long (0900-2300 hrs) and is stratified into two 7.0 hour time periods defined as A (0900-1600 hrs) and B (1601-2300 hrs). Additionally fishing days are stratified by type of day, i.e., weekend/holiday versus weekday. Days selected within any stratum represent the primary sampling units (of the two-stage design), and anglers interviewed represent the secondary sampling units. As opposed to the classic two-stage sampling design we do not know *a priori* the size of our secondary sampling units (i.e., the number of anglers available to sample on a

selected day-primary unit). Additionally, we will not be able to count all anglers returning on a selected sample day throughout the season. Accordingly, our variance estimation procedures will not involve the use of the within sample (between secondary unit-angler) variance component. However, because we will be interviewing the vast majority of all anglers in each selected sample, the finite population correction factor (fpc) associated with the secondary stage will be close to zero. The resulting within sample variance would make an essentially ignorable contribution to the overall variance estimate. This means our estimation procedure will collapse to a stratified random procedure, in which sample means (across all anglers interviewed within a sample) are used as the stratum observation.

The number of primary units to sample (i.e., days) was determined by using the simple random sampling design approach as outlined by Cochran (1977, section 4.6, pages 77-78). The sample size was selected using this approach rather than using a stratified approach, in that we do not have any good information regarding the variability of our desired estimates (harvest and catch per angler-day) on an individual stratum basis. However, we do have information from a previous study which indicate that the variability of harvest per unit effort (HPUE) of pink salmon, coho salmon, and Pacific halibut is on the order of a coefficient of variation (CV) of 40 to 75% for the entire season across all strata (Roth and Delaney, In Press). Using equation 4.5 from Cochran (1977, page 77) with a CV set to 60%, a relative precision set to ± 10%, and a t-value of 1.645 (associated with an α value of 0.10) the initial sample size is 101. After applying an fpc (equation 4.3, page 76 of Cochran 1977, with N = 186 possible samples = 2 possible samples each day times 93 days over the season), the resulting goal sample size is 66. This sample size should result in the desired precision listed in our objective criteria. Accordingly, we have scheduled a total of 67 samples (see below).

All weekend/holiday days and three of the five weekday days will be sampled each week. The weekdays not sampled were selected by randomly choosing one weekday and then randomly choosing the day before or after it also (in order to allow for two contiguous days-off for staff within each week). This procedure results in a constrained random sample of the weekday stratum. The resulting allocation proportions are as follows: 23.3% of the Mondays and Fridays, 16.7% of the Tuesdays and Thursdays, and 20% of the Wednesdays. Accordingly, samples collected on Mondays or Fridays will be weighted by a factor of 85.71% (resulting in a realized proportion of 20%); similarly samples collected on Tuesdays or Thursdays will be weighted by a factor of 120.00%. Samples collected on Wednesdays will not be weighted.

Within each component, two-thirds of the B periods were randomly selected for sampling, without replacement. Days not selected using this process were allocated to the A period. Allocation of sampling effort between the survey periods was based on the assumption that more effort will return during the evening than during the mornings within a day and that only one period could be sampled per day due to budget and personnel limits. The resultant sampling schedule for the survey is presented in Appendix A. The resulting sampling allocation is summarized as follows:

Type of Day	Number of Possible Samples	A Period Days Sampled #	A Period Days Sampled %	B Period Days Sampled #	B Period Days Sampled %	All Periods Days Sampled #	All Periods Days Sampled %	% of Possible Samples
Weekends	58	10	34.5	19	65.5	29	43.2	50.0
Weekdays	128	13	34.2	25	65.7	38	56.7	29.7
Total	186	23	34.3	44	65.6	67	100.0	36.0

One creel technician will perform the survey with assistance from the technician in charge of the marine catch sampling. The technician will conduct interviews of returning boat anglers that exit the fishery at the Valdez boat harbor. Individual anglers will be asked how many days they fished; the number of fish harvested, and the number released for each species (rockfish will not be segregated by species); and where they fished. All data will be collected on the marine survey form in accordance with specifications detailed in Appendix B. Anglers will also be given a questionnaire requesting their name and address for a follow-up survey (see Appendix B).

Whittier. A survey of the marine sport boat fishery operating out of Whittier will be conducted from 24 June through 4 September 1989.

At Whittier, the fishing day is defined as 12 hours long (1000-2200 hrs) and is stratified into four 3.0 hour time periods defined as A (1000-1300 hrs), B (1301-1600 hrs), C (1601-1900 hrs), and D (1901-2200 hrs). Accordingly, during the 73 days of the survey a total of 292 samples can be taken. Using the procedures outlined above for the Valdez survey, the goal sample size for the Whittier survey is 76. This sample size assumes a CV for HPUE in Whittier that is similar to the Valdez value. We do not have any similar information on the variability of HPUE in Whittier, but we expect the CV to be similar to that observed in the Valdez fishery. A total of 104 samples were selected (see below), accordingly we should obtain our desired objective criteria precision levels. This "over-sampling" should also protect us from a larger CV in Whittier (note, the sample size of 104 would be associated with a CV of 75%).

All weekend/holiday days and three of the five weekday days will be sampled each week. The weekdays not sampled were selected by randomly choosing one weekday and then randomly choosing the day before or after it also. Accordingly, each sampled weekday will be weighted during data analysis as noted above for the Valdez survey. On each day selected for sampling, one

period was selected for conducting the survey given the constraint that 50% of the sampling effort was assigned to period C, 25% to period D, 15% to period B, and 10% to A in each strata (weekday or weekend/holiday). Periods were randomly selected for sampling, without replacement. Allocation of sampling effort between the survey periods was based on discussions with the area manager of anticipated angler return patterns and constrained by the fact that only one period could be sampled per day due to budget and personnel limits. The resultant sampling schedule for the survey is presented in Appendix A. The sampling breakdown is as follows:

Type of Day	Number of Possible Samples	A Period Days Sampled #	A Period Days Sampled %	B Period Days Sampled #	B Period Days Sampled %	C Period Days Sampled #	C Period Days Sampled %	D Period Days Sampled #	D Period Days Sampled %	All Periods Days Sampled #	All Periods Days Sampled %	% of Possible Samples
Weekends	98	5	45.5	7	46.7	24	46.2	12	46.2	48	46.2	50.0
Weekdays	196	6	54.5	8	53.3	28	53.8	14	53.8	56	53.8	28.6
Total	292	11	10.6	15	14.4	52	50.0	26	25.0	104	100.0	35.6

One creel technician will perform the survey with assistance from the technician responsible for the marine catch sampling. Individual anglers will be asked how many days they fished; the number of fish caught, and the number released for each species (rockfish will not be segregated by species); and where they fished.

It is anticipated that most interviews will be obtained at the rail station as anglers depart Whittier via the train. Additional interviews will be obtained as anglers depart their boats at the docks. All data will be collected on the marine survey form in accordance with specifications detailed in Appendix B. Anglers will also be given a questionnaire requesting their name and address for a follow-up survey (see Appendix B).

Cordova. A survey of the marine sport boat fishery operating out of the Cordova boat harbor will be conducted from 24 June through 1 October 1989. The fishing day at Cordova is defined as 14 hours long (0800-2200) and is stratified into two 7.0 hour time periods defined as A (0800-1500) and B (1501-2200). Each 7 hour stratum was further subdivided into 3.5 hour sampling periods. As such, during the 100 days of the survey a total of 400 samples were possible. Accordingly, the goal sample size for this survey is 81 (assuming a CV of 60%, an α level of 0.10, and a relative precision of $\pm$ 10%), following the procedures used in the Valdez survey, as described above. The total number of samples allocated in the Cordova survey is 144, so we should obtain our desired objective criteria precision, even if the true CV is as high as 90%.

All weekend/holiday days and three weekdays will be sampled each week. The weekdays not sampled were selected by randomly choosing one weekday and then randomly choosing the day before or after it also. Accordingly, each sampled weekday will be weighted during data analysis as noted for the Valdez survey,

noted above. On each day selected for sampling, the creel technician will interview anglers for a total of 7 hours, divided into two 3.5 hour segments. (The length of time spent interviewing may need to be shortened by as much as an hour for both periods if it is found that the technician needs more time for catch sampling.) One quarter of the sampling effort was assigned to period A, and three quarters to period B. Periods were randomly selected for sampling without replacement. Allocation of sampling effort between the survey periods was based on the assumption that more anglers will return during the afternoon and evening than in the morning. The sampling breakdown is as follows:

Type of Day	Number of Possible Samples	A1 Period Days Sampled #	A1 Period Days Sampled %	A2 Period Days Sampled #	A2 Period Days Sampled %	B1 Period Days Sampled #	B1 Period Days Sampled %	B2 Period Days Sampled #	B2 Period Days Sampled %	All Periods Days Sampled #	All Periods Days Sampled %	% of Possible Samples
Weekends	128	5	27.8	11	52.4	25	44.6	23	46.9	64	44.4	50.0
Weekdays	272	13	72.2	10	47.6	31	55.4	26	53.1	80	55.6	29.4
Total	400	18	12.5	21	14.6	56	38.9	49	34.0	144	100.0	36.0

One technician will perform the survey and do marine catch sampling. Individual anglers will be asked how many days they fished; the number of fish caught, and the number released for each species (rockfish will not be segregated by species); and where they fished. All data will be collected on the marine survey form in accordance with specifications detailed in Appendix B. Anglers will also be given a questionnaire requesting their name and address for a follow-up survey (see Appendix B).

Homer. A survey of the marine sport boat fishery operating out of the Homer boat harbor will be conducted from 1 July through 1 September 1989. The fishing day for Homer is defined as 15.5 hours long (0630-2200), and is stratified into four periods: A (0630-1000), B (1001-1330), C (1331-1700), and D (1701-2200). Accordingly, a total of 252 possible samples were available over the 63 days of the survey. The resultant sample size goal is 73 (using procedures similar to those described in the Valdez survey, above). The total number of allocated samples is 88 (see below), so we should obtain our desired objective criteria precision levels.

All weekend/holiday days and three weekdays will be sampled each week. The weekdays not sampled were selected by randomly choosing one weekday and then randomly choosing the day before or after it also. Accordingly, each sampled weekday will be weighted during data analysis as noted for the Valdez survey, noted above. Ten percent of the sampling effort was assigned to period A, 25% to period B, 40% to period C, and 25% to period D. Periods were randomly selected for sampling without replacement. During period C one technician will interview returning charter boat anglers and the other will interview private boats. During all other periods the creel technician will attempt to interview an equal proportion of charter boat and private boat anglers. Allocation of sampling effort between the survey periods was based on the assumption that most anglers will return during period C, with few returning

in A and, moderate numbers returning in B and D. The sampling breakdown is as follows:

Type of Day	Number of Possible Samples	A Period Days Sampled #	A Period Days Sampled %	B Period Days Sampled #	B Period Days Sampled %	C Period Days Sampled #	C Period Days Sampled %	D Period Days Sampled #	D Period Days Sampled %	All Periods Days Sampled #	All Periods Days Sampled %	% of Possible Samples
Weekends	76	5	41.7	10	43.5	13	43.3	10	43.5	38	43.2	50.0
Weekdays	176	7	58.3	13	56.5	17	56.7	13	56.5	50	56.8	28.4
Total	252	12	13.6	23	26.1	30	34.1	23	26.1	88	100.0	34.9

One technician will perform the survey with the assistance of the technician in charge of marine catch sampling. Individual anglers will be asked how many days they fished; the number of fish caught by species, and the number released for each species (rockfish will not be segregated by species); and where they fished. All data will be collected on the marine survey form in accordance with specifications detailed in Appendix B. Anglers will also be given a questionnaire requesting their name and address for a follow-up survey (see Appendix B).

Kodiak. The survey of the marine sport boat fishery operating out of the Kodiak boat harbor will be conducted from 1 July through 15 September 1989. The fishing day for Kodiak is defined as 13 hours long (1000-2300), and is stratified into four periods: A (1000-1400), B (1401-1700), C (1701-2000), and D (2001-2300). Accordingly, a total of 308 possible samples were available over the 77 days of the survey. The resultant sample size goal is 77 (using procedures similar to those described in the Valdez survey, above). The total number of samples allocated in the Kodiak survey is 106, so we should obtain our desired objective criteria precision, even if the true CV is as high as 75%.

All weekend/holiday days and three weekdays will be sampled each week. The weekdays not sampled were selected by randomly choosing one weekday and then randomly choosing the day before or after it also. Accordingly, each sampled weekday will be weighted during data analysis as noted for the Valdez survey, noted above. Twenty percent of the sampling effort was assigned to period A, 30% to period B, 30% to period C, and 20% to period D. The allocation of sampling effort between the survey periods was based on the area management biologist's best guess at the percent of anglers returning during each period. The sampling breakdown is as follows:

Type of Day	Number of Possible Samples	A Period Days Sampled #	A Period Days Sampled %	B Period Days Sampled #	B Period Days Sampled %	C Period Days Sampled #	C Period Days Sampled %	D Period Days Sampled #	D Period Days Sampled %	All Periods Days Sampled #	All Periods Days Sampled %	% of Possible Samples
Weekends	96	9	45.0	15	45.5	15	45.5	9	45.0	48	45.3	50.0
Weekdays	212	11	55.0	18	54.5	18	54.5	11	55.0	58	54.7	27.4
Total	308	20	18.9	33	31.1	33	31.1	20	18.9	106	100.0	34.4

One technician will perform the survey with the assistance of the technician in charge of marine catch sampling. Individual anglers will be asked how many days they fished; the number of fish caught, and the number released for each species (rockfish will not be segregated by species); and where they fished. All data will be collected on the marine survey form in accordance with specifications detailed in Appendix B. Anglers will also be given a questionnaire requesting their name and address for a follow-up survey (see Appendix B).

Data Reduction and Analysis:

Mark sense forms will be visually examined for errors by the technicians and then sent to the Division of Sport Fish's Research and Technical Services group (RTS). RTS will optically scan the mark sense forms to produce a computer data file, a frequency report, and a listing of each batch of data. The file and reports will be returned to the technician for editing and analysis. The final, edited data file will be returned to RTS for permanent storage on tape.

The mean effort and mean harvest by interviewed anglers will be estimated for the weekday and weekend/holiday components of each fishery. The variance of these means will be estimated using a two-stage sample design (Von Geldern and Tomlinson 1973) with days being considered the primary sample units (which there are a finite number available) and anglers the secondary sample units. There are an unknown number of anglers available to sample each day. As noted above, our variance estimation procedures will <u>not</u> involve the use of the within sample (between secondary unit-angler) variance component. However, because we will be interviewing the vast majority of all anglers in each selected sample, the finite population correction factor (fpc) associated with the secondary stage will be close to zero. The resulting within sample

variance would make an essentially ignorable contribution to the overall variance estimate. This means our estimation procedure will collapse to a stratified random procedure, in which sample means (across all anglers interviewed within a sample) are used as the stratum observation.

Catch and harvest per angler-day will be estimated for each day as the quotient of the mean harvest and mean effort estimates given the assumption that there are insignificant differences in rates between periods within a day. Accordingly, only the type of fishing day will be used to define the stratum during the data analysis phase. The variance of the catch and harvest rate will be estimated using the approximation for the quotient of the means of two random variables (Jessen 1978, equation 5.8, page 128).

Names and addresses of persons willing to participate in a follow-up survey will be forwarded to the economic study group.

Creel Surveys

Effort, catch and harvest rates, and selected characteristics of anglers participating in two sport fisheries near Cordova and the sport fishery at Eshamy Lagoon will be estimated using stratified two-stage creel surveys. The two creel surveys near Cordova are: (1) a survey of two roadside sport fisheries of the Copper River Delta (henceforward referred to as the Delta fishery), and (2) a survey of the sport fishery near Eyak Lake (henceforward referred to as the Eyak fishery).

Study Design and Data Collection:

For all creel surveys, the number of primary units to sample was determined by using the simple random sampling design approach as outlined by Cochran (1977, section 4.6, pages 77-78). The sample size was selected using this approach rather than using a stratified approach, in that we do not have any good information (at this time) regarding the variability of our desired estimates (effort, harvest, and catch) on an individual stratum basis. However, we do have information from previous studies which indicate that the approximate variability of our estimates. The individual details for each survey is outlined in each section below.

Delta Fishery. The creel survey of the Delta fishery will be conducted from 15 June through 1 October 1989. Two areas in the Delta will be surveyed: the Alaganik River and Clear Creek.

Effort, catch and harvest rates in the Delta fishery will be estimated using a stratified random creel survey. Effort and catch and harvest rate will be estimated separately for weekday and weekend/holiday strata. The fishing day is defined as 14.5 hours long (0600-2030 hrs) and is stratified into three unequal time periods defined as A (0600-0930 hrs), B (0931-1700 hrs), and C (1701-2030 hrs) based on discussions with the area manager regarding anticipated angler use patterns.

Similar surveys of fisheries of approximately the same size indicate that angler catch is estimated with an approximate CV of 65% (note that angler effort is usually estimated more precisely in this type of survey, so we use angler catch for sample size determination). Using equation 4.5 from Cochran (1977, page 77) with a CV set to 65%, a relative precision set to ± 10%, and a t-value of 1.96 (associated with an α value of 0.05) the initial sample size is 170. After applying an fpc (equation 4.3, page 76 of Cochran 1977, with N = 654 possible samples = 6 possible samples each day times 109 days over the season), the resulting goal sample size is 135. This sample size should result in the desired precision listed in our objective criteria. Accordingly, we have scheduled a total of 138 samples (see below).

All weekend/holiday days and three of the five weekday days will be sampled each week. The weekdays not sampled were selected by randomly choosing one weekday and then randomly choosing the day before or after it also. Accordingly, weekday samples will be weighted as outlined above for the marine surveys.

Within each stratum, periods were randomly selected for sampling, without replacement given the constraint that only one B period or one each A and C period could be sampled in a day. For the weekday strata, allocation of sampling effort between periods was as follows: 40% each of the available samples for periods A and C and 20% for period B. For the weekend/holiday strata, allocation of sampling effort between periods was as follows: 35% each for periods A and C and 30% for period B. Allocation of sampling effort between the survey periods during the weekday strata was based on the assumption that more effort will occur during the mornings and evenings than during the midday period. Allocation of sampling effort between the survey periods during the weekend/holiday strata was based on the assumption that effort will occur regardless of time of day. An additional constraint was that only period B or periods A and C could be sampled each day due to budget and personnel limits. The resultant sampling schedule for the survey is presented in Appendix A. The sampling breakdown is as follows:

Type of Day	Number of Possible Samples	A Period Days Sampled #	A Period Days Sampled %	B Period Days Sampled #	B Period Days Sampled %	C Period Days Sampled #	C Period Days Sampled %	All Periods Days Sampled #	All Periods Days Sampled %	% of Possible Samples
Weekends	210	25	42.4	10	50.0	25	42.4	60	43.5	58.6
Weekdays	444	34	57.6	10	50.0	34	57.6	78	56.5	17.6
Total	654	59	42.8	20	14.4	59	42.8	138	100.0	21.1

Since two areas are to be surveyed, during a selected A or C period 1.5 hours will be spent at each area and during a selected B period 3.5 hours will be spent at each area. The areas will be surveyed in random order determined prior to each sample period.

Counts of anglers will be used to estimate fishing effort in units of angler-hours and interviews of anglers will be used to estimate catch and harvest rates (number of fish per hour). One technician will perform the survey. The technician will perform a count of anglers during a randomly selected 15-minute interval during the daily sampling period. Counts will be considered instantaneous and representative of the effort during that period (Neuhold and Lu 1957). During the count, the technician will count all anglers actively fishing.

During the remaining time in the period, the technician will conduct interviews of individual anglers. Individual anglers will be asked how long they fished, the number by species they caught and the number by species they kept. All fish in their possession will be inspected for tags and will be measured. All interviewed anglers will be given a questionnaire requesting their name and address for a follow-up survey (see Appendix B). As many completed-trip anglers will be interviewed as possible; however, it is expected that the majority of the interviews will be incomplete-trip interviews. All data will be collected on the marine interview mark-sense form in accordance with specifications detailed in Appendix B.

Eyak Fishery. The creel survey of the Eyak fishery will be conducted from 15 June through 1 October 1989.

Effort, catch and harvest rates during the sport fishery near Eyak Lake will be estimated using a stratified random creel survey. Effort and catch and harvest rate will be estimated separately for weekday and weekend/holiday strata. The fishing day is defined as 14.5 hours long (0600-2030 hrs) and is stratified into three unequal time periods defined as A (0600-0930 hrs), B (0931-1700 hrs), and C (1701-2030 hrs), based on discussions with the area manager regarding anticipated angler use patterns.

As noted above, similar surveys of fisheries of approximately the same size indicate that angler catch is estimated with an approximate CV of 65% . Using equation 4.5 from Cochran (1977, page 77) with a CV set to 65%, a relative precision set to ± 10%, and a t-value of 1.96 (associated with an α value of 0.05) the initial sample size is 170. After applying an fpc (equation 4.3, page 76 of Cochran 1977, with N = 327 possible samples = 3 possible samples each day times 109 days over the season), the resulting goal sample size is 112. This sample size should result in the desired precision listed in our objective criteria. Accordingly, we have scheduled a total of 139 samples, so that we could experience a CV as large as 75% and still obtain our desired precision.

All weekend/holiday days and three of the five weekday days will be sampled each week. The weekdays not sampled were selected by randomly choosing one weekday and then randomly choosing the day before or after it also. Accordingly, each sampled weekday will be weighted during data analysis as noted above for the Delta fishery survey. Within each strata, periods were randomly selected for sampling, without replacement, given the constraint that only one B period or one each A and C period could be sampled in a day. For the weekday strata, allocation of sampling effort between periods was as follows: 40% each for periods A and C and 20% for period B. For the

weekend/holiday strata, allocation of sampling effort between periods was as follows: 35% each for periods A and C and 30% for period B. Allocation of sampling effort between the survey periods during the weekday strata was based on the assumption that more effort will occur during the mornings and evenings than during the midday period. Allocation of sampling effort between the survey periods during the weekend/holiday strata was based on the assumption that effort will occur regardless of time of day. An additional constraint was that only period B or periods A and C could be sampled each day due to budget and personnel limits. The resultant sampling schedule for the survey is presented in Appendix A. The sampling breakdown is as follows:

Type of Day	Number of Possible Samples	A Period Days Sampled #	A Period Days Sampled %	B Period Days Sampled #	B Period Days Sampled %	C Period Days Sampled #	C Period Days Sampled %	All Periods Days Sampled #	All Periods Days Sampled %	% of Possible Samples
Weekends	105	25	41.7	10	52.6	25	41.7	60	43.2	57.1
Weekdays	222	35	58.3	9	47.4	35	58.3	79	56.8	35.6
Total	327	60	43.2	19	13.6	60	43.2	139	100.0	42.5

Counts of anglers will be used to estimate fishing effort in units of angler-hours and interviews of anglers will be used to estimate catch and harvest rates (number of fish per hour) for each species of interest during the survey at Eyak Lake. The technician will perform a count of anglers during a randomly selected 15-minute interval during the sampling period. Counts will be considered instantaneous and representative of the effort during that period (Neuhold and Lu 1957). During the count, the technician will count all anglers actively fishing. During the remaining time in the period, the technician will conduct interviews of individual anglers. Individual anglers will be asked how long they fished, the number by species they caught and the number by species they kept. All fish in their possession will be inspected for tags and will be measured. All interviewed anglers will be given a questionnaire requesting their name and address for a follow-up survey (see s ppendix B). As many completed-trip anglers will be interviewed as possible; however, it is expected that the majority of the interviews will be incomplete-trip interviews. All data will be collected on the marine interview mark-sense form in accordance with specifications detailed in Appendix B.

Eshamy Lagoon. A creel survey of the Eshamy sport fishery will be conducted from 1 July through 4 September 1989.

Effort, catch and harvest rates during the sport fishery in Eshamy Lagoon will be estimated using a stratified random creel survey. Effort and catch and harvest rate will be estimated separately for weekday and weekend/holiday strata. The fishing day is 14 hours long (0800-2200 hrs) and is divided into four 3.5 hour time periods defined as A: 0800-1130 hrs, B: 1131-1500 hrs, C: 1501-1830 hrs, and D: 1831-2200 hrs. Periods are stratified based on stage as affected by the tide. Periods during which a select high stage occurs are

categorized as high use stage periods. All other periods are categorized as low use stage periods.

A similar survey conducted in 1988 indicated that the overall CV for estimating angler catch in this fishery was approximately 25%. Using equation 4.5 from Cochran (1977, page 77) with a CV set to 25%, a relative precision set to = 7.5%, and a t-value of 1.96 (associated with an a value of 0.05) the initial sample size is 46. After applying an fpc (equation 4.3, page 76 of Cochran 1977, with N = 264 possible samples = 4 possible samples each day times 66 days over the season), the resulting goal sample size is 40. This sample size should result in the desired precision listed in our objective criteria. Accordingly, we have scheduled a total of 49 samples, so that we could experience a larger CV than the planned 25% level.

All weekend/holiday days and three of the five weekday days will be sampled each week. The weekdays not sampled were selected by randomly choosing one weekday and then randomly choosing the day before or after it also. Accordingly, each sampled weekday will be weighted during data analysis as noted above for the Delta fishery survey. On each day selected for sampling, one period was selected for conducting the survey, given the constraint that 75% of the high use stage periods were selected in each strata (weekday or weekend/holiday). Allocation of sampling effort between the survey periods was based on the assumption that more effort will occur during high use stages than during other tidal stages within a day and that only one period could be sampled per day due to budget and personnel limits. The resultant sampling schedule for the survey is presented in Appendix A. The sampling breakdown is as follows:

Type of Day	Number of Possible Samples	A Period Days Sampled #	A Period Days Sampled %	B Period Days Sampled #	B Period Days Sampled %	C Period Days Sampled #	C Period Days Sampled %	D Period Days Sampled #	D Period Days Sampled %	All Periods Days Sampled #	All Periods Days Sampled %	% of Possible Samples
Weekends	92	2	50.0	10	58.8	10	40.0	1	12.5	23	46.9	25.0
Weekdays	172	2	50.0	7	41.2	10	50.0	7	87.5	26	53.1	15.1
Total	264	4	18.9	17	31.1	20	31.1	8	18.9	49	100.0	18.6

The sampling breakdown according to stage use periods follows:

Type of Day	Number of Possible Samples	High Use Stage Periods Sampled #	High Use Stage Periods Sampled %	Low Use Stage Periods Sampled #	Low Use Stage Periods Sampled %	All Periods Days Sampled #	All Periods Days Sampled %	% of Possible Samples
Weekends	92	18	51.4	5	35.7	23	46.9	25.0
Weekdays	172	17	48.6	9	64.3	26	53.1	15.1
Total	264	35	71.4	14	28.6	49	100.0	36.0

Counts of anglers will be used to estimate fishing effort in units of angler-hours, and interviews of anglers will be used to estimate catch and harvest rates (number of fish per hour). One technician will perform the survey. The technician will perform a count of anglers during a randomly selected 15-minute interval during the daily sampling period. Counts will be considered instantaneous and representative of the effort during that period (Neuhold and Lu 1957). During the count, the technician will count all anglers actively fishing.

During the remaining time in the period, the technician will conduct interviews of individual anglers. Individual anglers will be asked how long they fished, the number by species they caught and the number by species they kept. All fish in their possession will be inspected for tags and will be measured. All interviewed anglers will be given a questionnaire requesting their name and address for a follow-up survey (see Appendix B). As many completed-trip anglers will be interviewed as possible; however, it is expected that the majority of the interviews will be incomplete-trip interviews. All data will be collected on the marine interview mark-sense form in accordance with specifications detailed in Appendix B.

Data Reduction and Analysis:

Mark sense forms will be visually examined for errors by the technicians and then sent to the Division of Sport Fish's Research and Technical Services group (RTS). RTS will optically scan the mark sense forms to produce a computer data file, a frequency report, and a listing of each batch of data. The file and reports will be returned to the technician for editing and analysis. The final, edited data file will be returned to RTS for permanent storage on tape.

Effort in angler-hours will be estimated separately for the weekday and weekend/holiday strata for each area. The estimates for Eshamy Lagoon will be further stratified by high use stage vs low use stages. A stratified random sample design will be used to estimate effort and its variance (Scheaffer et al. 1979). All estimates are considered independent estimates, therefore, seasonal estimates of effort and variance are the sum of these quantities over all strata. Samples collected on the weekdays will be weighted as noted above. Angler effort is estimated by multiplying the average angler count obtained over all samples within a stratum by the hours available for fishing in that stratum. The effort estimates and associated variance estimates are obtained following the approach outlined by Von Geldern and Tomlinson (1973).

Angler catch and harvest rates (number of fish per hour) of each species will be estimated separately for the weekday and weekend/holiday strata (and use-stage strata for Eshamy) for each area from both completed-trip and incompleted-trip interview data. If sufficient completed-trip data are obtained, the catch and harvest rates of the completed-trip and incompleted-trip anglers will be tested for differences, by strata and area, using a sign test (Daniel 1978, pages 27-28). If a difference is detected, then only

completed-trip data will be used to estimate catch and harvest rates. If there are not sufficient completed-trip data, it will be assumed that the incompleted-trip data provides an unbiased estimate of completed-trip information. The variances of the mean effort per angler and mean catch and harvest per angler will be estimated using a two-stage sample design (Von Geldern and Tomlinson 1973) with days being considered the primary sample units (which there are a finite number available) and anglers being considered the secondary sample units (which there are an unknown number available to sample each day). Catch and harvest rates will be estimated for each day as the quotient of the mean catch or harvest and the mean effort, given the assumption that there is no significant difference in catch or harvest rates between periods in any day. The variance of these rates will be estimated using the approximation for the quotient of the means of two random variables (Jessen 1978, equation 5.8, page 128).

The catch and harvest of each species during the weekday and weekend/holiday strata (and use-stage strata for Eshamy) for each area will be estimated as the product of the estimated effort (angler-hours) and catch/harvest rate (number of fish per angler-hour). Variance will be estimated using Goodman's (1960) formula for the exact variance of the product of two independent random variables. All strata estimates are considered independent estimates, therefore, seasonal estimates and their variances are the sum of these quantities over all strata.

Anchorage Float-plane Logbook Survey

A logbook survey of anglers sport anglers who access PWS from the float-plane base in Anchorage will be conducted to collect data on selected demographic characteristics and general catch and effort parameters.

The logbook survey of Anchorage based charter aircraft operators will be conducted from 15 June to 15 September 1989.

Days fished, area fished, catch, and harvest by anglers who travel to PWS via air charter out of Anchorage will be estimated using a logbook survey. All major air charter operators out of Anchorage will be asked to keep a log for each charter they conduct to PWS. A copy of the log along with instructions for completing it are presented in Appendix C.

Anglers contacted in this survey will also be given the Oil Spill Impact Assessment Information Form to ask for their name and address if they are willing to respond to a follow-up questionnaire.

When the log is initially given to the air charter operators, the instructions for completing it will be reviewed and any questions they may have answered by a member of the Anchorage Sport Fish Division staff assigned to this survey. They will be also given the telephone number of the Anchorage Fish and Game office to call if any questions may arise. During the early part of the season, the same member of the Anchorage staff will work with the air charter operators to assure that the log is being correctly completed. All logs will be picked up biweekly to prevent loss and check for data integrity.

SCHEDULES AND REPORTS

A schedule of tasks to be completed is presented as follows:

Task	Dates
Valdez Marine Surveys	15 June - 15 Sept. 1989
Whittier Marine Surveys	24 June - 04 Sept. 1989
Cordova Marine Surveys	24 June - 01 Oct. 1989
Homer Marine Surveys	1 July - 1 Sept. 1989
Kodiak Marine Surveys	1 July - 15 Sept. 1989
Cordova Creel Surveys	15 June - 01 Oct. 1989
Eshamy Creel Surveys	01 July - 04 Sept. 1989
Anchorage Operations	15 June - 15 Sept. 1989
Marine Surveys opscanned	October 1989
Marine Survey tabulation	January 1989
Creel Survey Data Reduction and Analysis	December 1989
Draft Report	February 1990
Draft FY91 Operational Plan	March 1990

Results of these study efforts will be reported to the Division of Oil Spill Impact Assessment and Restoration. Upon completion of litigative concerns, these data will be published as either an Alaska Department of Fish and Game, Sport Fish Division, Fishery Data Series report or in the fisheries literature.

RESPONSIBILITIES

Craig Whitmore, Fisheries Biologist III - Project Leader

> Duties: Supervision of all aspects of the project. Drafting of operational plans, development of sampling methods and procedures. Post-seasonally, will be responsible for recorded data analysis and reporting of results.

Kent Roth, Fisheries Biologist II - Assistant Project Leader

> Duties: Supervision of all aspects of the project. Drafting of operational plans, development of sampling methods and procedures. Post-seasonally, will be responsible for recorded data analysis and reporting of results.

Wilson Potterville, Fisheries Biologist II

> Duties: Coordination of sampling schedules and supervision of project personnel in Valdez and Cordova. Post season support for recorded data analysis and reporting of results in the Fisheries Data Series report.

John B. Murray, Fisheries Biologist III

Duties: Coordination of sampling schedules and supervision of project personnel in Kodiak.

Fishery Technicians

Duties: Collection of the field data. Will assist in preliminary analysis of the recorded data.

BUDGET SUMMARY

A detailed budget[1] for the 1989 oil year is:

Line Item	Category	Budget
100	Personnel Services	$ 84,500
200	Travel	$ 7,000
300	Contractual	$ 54,900
400	Commodities	$ 9,500
500	Equipment	$ 20,000
700	Grants	$ 0
Total		$ 175,900

[1] Budget is for all activities performed from March 27, 1989 to February 28, 1990.

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APPENDICES

APPENDIX

A. Creel Survey Schedules

Sampling schedule for the Valdez Bay boat sport fishery, 1989.

Date	Strata[1]	Period/Time A:0900-1600	Period/Time B:1601-2300
15-Jun	WD	X	
16-Jun	WD		X
17-Jun	WE		X
18-Jun	WE		X
19-Jun	WD	X	
20-Jun	WD		
21-Jun	WD		
22-Jun	WD		X
23-Jun	WD	X	
24-Jun	WE	X	
25-Jun	WE	X	
26-Jun	WD	X	
27-Jun	WD	X	
28-Jun	WD		X
29-Jun	WD		
30-Jun	WD		
01-Jul	WE	X	
02-Jul	WE		X
03-Jul	WE		X
04-Jul	WE	X	
05-Jul	WD		
06-Jul	WD		
07-Jul	WD		X
08-Jul	WE		X
09-Jul	WE		X
10-Jul	WD		X
11-Jul	WD	X	
12-Jul	WD		X
13-Jul	WD		
14-Jul	WD		
15-Jul	WE	X	
16-Jul	WE		X
17-Jul	WD		X
18-Jul	WD		X
19-Jul	WD		X
20-Jul	WD		
21-Jul	WD		
22-Jul	WE		X
23-Jul	WE	X	
24-Jul	WD		X
25-Jul	WD	X	

-Continued-

Sampling schedule for the Valdez Bay boat sport fishery, 1989 (continued).

Date	Strata[1]	Period/Time A:0900-1600	B:1601-2300
26-Jul	WD		
27-Jul	WD		
28-Jul	WD	X	
29-Jul	WE	X	
30-Jul	WE		X
31-Jul	WD		
01-Aug	WD		
02-Aug	WD		X
03-Aug	WD		X
04-Aug	WD		X
05-Aug	WE		X
06-Aug	WE		X
07-Aug	WD		X
08-Aug	WD	X	
09-Aug	WD		
10-Aug	WD		
11-Aug	WD		X
12-Aug	WE	X	
13-Aug	WE		X
14-Aug	WD	X	
15-Aug	WD		X
16-Aug	WD		
17-Aug	WD		
18-Aug	WD		X
19-Aug	WE	X	
20-Aug	WE		X
21-Aug	WD	X	
22-Aug	WD		X
23-Aug	WD		
24-Aug	WD		
25-Aug	WD		X
26-Aug	WE		X
27-Aug	WE		X
28-Aug	WD		X
29-Aug	WD	X	
30-Aug	WD		X
31-Aug	WD		
01-Sep	WD		
02-Sep	WE		X
03-Sep	WE		X
04-Sep	WE		X

-Continued-

Sampling schedule for the Valdez Bay boat sport fishery, 1989 (continued).

Date	Strata[1]	Period/Time A:0900-1600	Period/Time B:1601-2300
05-Sep	WD		X
06-Sep	WD	X	
07-Sep	WD		
08-Sep	WD		
09-Sep	WE		X
10-Sep	WE	X	
11-Sep	WD		
12-Sep	WD		
13-Sep	WD		X
14-Sep	WD		X
15-Sep	WD		X

[1] WD=Weekday, WE=Weekend/Holiday.

Whittier creel survey schedule, 1989.

DATE	DAY	A (1000-1300)	B (1301-1600)	C (1601-1900)	D (1901-2200)
JUNE 24	SAT			X	X
JUNE 25	SUN		X	X	
JUNE 26	MON			X	X
JUNE 27	TUES			X	X
JUNE 28	WEDS	X		X	
JUNE 29	THURS	----------OFF----------			
JUNE 30	FRI	----------OFF----------			
JULY 01	SAT			X	X
JULY 02	SUN			X	X
JULY 03	MON		X	X	
JULY 04	TUES			X	X
JULY 05	WEDS	----------OFF----------			
JULY 06	THURS	----------OFF----------			
JULY 07	FRI	----------OFF----------			
JULY 08	SAT		X	X	
JULY 09	SUN			X	X
JULY 10	MON	----------OFF----------			
JULY 11	TUES	----------OFF----------			
JULY 12	WEDS		X	X	
JULY 13	THURS		X	X	
JULY 14	FRI			X	X
JULY 15	SAT	X		X	
JULY 16	SUN			X	X
JULY 17	MON	----------OFF----------			
JULY 18	TUES	----------OFF----------			
JULY 19	WEDS		X	X	
JULY 20	THURS	X		X	
JULY 21	FRI	X		X	
JULY 22	SAT		X	X	
JULY 23	SUN		X	X	
JULY 24	MON	----------OFF----------			
JULY 25	TUES	----------OFF----------			
JULY 26	WEDS			X	X
JULY 27	THURS			X	X
JULY 28	FRI			X	X
JULY 29	SAT	X		X	
JULY 30	SUN	X		X	
JULY 31	MON		X	X	

-continued-

Whittier creel survey schedule, 1989 (continued).

DATE	DAY	A (1000-1300)	B (1301-1600)	C (1601-1900)	D (1901-2200)
AUG 01	TUES		X	X	
AUG 02	WEDS			X	X
AUG 03	THUR	----------OFF----------			
AUG 04	FRI	----------OFF----------			
AUG 05	SAT			X	X
AUG 06	SUN		X	X	
AUG 07	MON	----------OFF----------			
AUG 08	TUES	----------OFF----------			
AUG 09	WEDS			X	X
AUG 10	THUR		X	X	
AUG 11	FRI		X	X	
AUG 12	SAT		X	X	
AUG 13	SUN			X	X
AUG 14	MON	X		X	
AUG 15	TUES			X	X
AUG 16	WEDS	----------OFF----------			
AUG 17	THUR	----------OFF----------			
AUG 18	FRI			X	X
AUG 19	SAT			X	X
AUG 20	SUN		X	X	
AUG 21	MON	X		X	
AUG 22	TUES	X		X	
AUG 23	WEDS			X	X
AUG 24	THUR	----------OFF----------			
AUG 25	FRI	----------OFF----------			
AUG 26	SAT			X	X
AUG 27	SUN	X		X	
AUG 28	MON			X	X
AUG 29	TUES	----------OFF----------			
AUG 30	WEDS	----------OFF----------			
AUG 31	THUR			X	X
SEPT 01	FRI			X	X
SEPT 02	SAT			X	X
SEPT 03	SUN			X	X
SEPT 04	MON	X		X	

Cordova Boat Harbor Creel Survey Schedule, 1989

DATE	DAY	A1 (0800-1130)	A2 (1131-1500)	B1 (1501-1830)	B2 (1831-2200)
JUNE 24	SAT			X	X
JUNE 25	SUN			X	X
JUNE 26	MON	X		X	
JUNE 27	TUES		X	X	
JUNE 28	WEDS			X	X
JUNE 29	THURS	----------	DAY OFF	----------	
JUNE 30	FRI	----------	DAY OFF	----------	
JULY 01	SAT		X		X
JULY 02	SUN			X	X
JULY 03	MON	----------	DAY OFF	----------	
JULY 04	TUES	X		X	
JULY 05	WEDS	----------	DAY OFF	----------	
JULY 06	THURS	----------	DAY OFF	----------	
JULY 07	FRI			X	X
JULY 08	SAT			X	X
JULY 09	SUN			X	X
JULY 10	MON	----------	DAY OFF	----------	
JULY 11	TUES	----------	DAY OFF	----------	
JULY 12	WEDS		X	X	
JULY 13	THURS		X	X	
JULY 14	FRI			X	X
JULY 15	SAT		X	X	
JULY 16	SUN			X	X
JULY 17	MON			X	X
JULY 18	TUES		X	X	
JULY 19	WEDS	----------	DAY OFF	----------	
JULY 20	THURS	----------	DAY OFF	----------	
JULY 21	FRI			X	X
JULY 22	SAT			X	X
JULY 23	SUN	X	X		
JULY 24	MON			X	X
JULY 25	TUES			X	X
JULY 26	WEDS	----------	DAY OFF	----------	
JULY 27	THURS	----------	DAY OFF	----------	
JULY 28	FRI		X		X
JULY 29	SAT	X		X	
JULY 30	SUN	X			X
JULY 31	MON		X	X	

-continued-

Cordova Boat Harbor Creel Survey Schedule, 1989 (continued).

DATE	DAY	A1 (0800-1130)	A2 (1131-1500)	B1 (1501-1830)	B2 (1831-2200)
AUG 01	TUES			X	X
AUG 02	WEDS	X			X
AUG 03	THUR	----------	DAY OFF	----------	
AUG 04	FRI	----------	DAY OFF	----------	
AUG 05	SAT			X	X
AUG 06	SUN			X	X
AUG 07	MON			X	X
AUG 08	TUES		X		X
AUG 09	WEDS			X	X
AUG 10	THUR	----------	DAY OFF	----------	
AUG 11	FRI	----------	DAY OFF	----------	
AUG 12	SAT			X	X
AUG 13	SUN			X	X
AUG 14	MON			X	X
AUG 15	TUES	----------	DAY OFF	----------	
AUG 16	WEDS	----------	DAY OFF	----------	
AUG 17	THUR	X	X		
AUG 18	FRI	X			X
AUG 19	SAT		X	X	
AUG 20	SUN		X	X	
AUG 21	MON			X	X
AUG 22	TUES			X	X
AUG 23	WEDS	----------	DAY OFF	----------	
AUG 24	THUR	----------	DAY OFF	----------	
AUG 25	FRI	X		X	
AUG 26	SAT	X		X	
AUG 27	SUN		X		X
AUG 28	MON			X	X
AUG 29	TUES		DAY OFF		
AUG 30	WEDS		DAY OFF		
AUG 31	THUR			X	X

-continued-

Cordova Boat Harbor Creel Survey Schedule, 1989 (continued).

DATE	DAY	A1 (0800-1130)	A2 (1131-1500)	B1 (1501-1830)	B2 (1831-2200)
SEPT 01	FRI	X		X	
SEPT 02	SAT			X	X
SEPT 03	SUN			X	X
SEPT 04	MON		X		X
SEPT 05	TUES		X		X
SEPT 06	WEDS	----------	DAY OFF	----------	
SEPT 07	THURS	----------	DAY OFF	----------	
SEPT 08	FRI	----------	DAY OFF	----------	
SEPT 09	SAT			X	X
SEPT 10	SUN			X	X
SEPT 11	MON			X	X
SEPT 12	TUES	----------	DAY OFF	----------	
SEPT 13	WEDS	----------	DAY OFF	----------	
SEPT 14	THURS	X		X	
SEPT 15	FRI			X	X
SEPT 16	SAT			X	X
SEPT 17	SUN		X	X	
SEPT 18	MON	----------	DAY OFF	----------	
SEPT 19	TUES	----------	DAY OFF	----------	
SEPT 20	WEDS			X	X
SEPT 21	THURS	X			X
SEPT 22	FRI	X		X	
SEPT 23	SAT			X	X
SEPT 24	SUN		X		X
SEPT 25	MON	----------	DAY OFF	----------	
SEPT 26	TUES	----------	DAY OFF	----------	
SEPT 27	WEDS		X	X	
SEPT 28	THURS			X	X
SEPT 29	FRI			X	X
SEPT 30	SAT		X	X	
OCT 01	SUN		X		X

Homer creel survey schedule, 1989.

DATE	DAY	A (1000-1300)	B (1301-1600)	C (1601-1900)	D (1901-2200)
JULY 01	SAT		X	X	
JULY 02	SUN	X		X	
JULY 03	MON			X	X
JULY 04	TUES		X		X
JULY 05	WEDS	----------OFF----------			
JULY 06	THURS	----------OFF----------			
JULY 07	FRI	----------OFF----------			
JULY 08	SAT			X	X
JULY 09	SUN		X		X
JULY 10	MON	X			X
JULY 11	TUES		X	X	
JULY 12	WEDS	X		X	
JULY 13	THURS	----------OFF----------			
JULY 14	FRI	----------OFF----------			
JULY 15	SAT			X	X
JULY 16	SUN		X	X	
JULY 17	MON		X	X	
JULY 18	TUES	----------OFF----------			
JULY 19	WEDS	----------OFF----------			
JULY 20	THURS	X	X		
JULY 21	FRI		X		X
JULY 22	SAT	X	X		
JULY 23	SUN			X	X
JULY 24	MON	----------OFF----------			
JULY 25	TUES	----------OFF----------			
JULY 26	WEDS		X	X	
JULY 27	THURS			X	X
JULY 28	FRI			X	X
JULY 29	SAT		X	X	
JULY 30	SUN	X			X
JULY 31	MON		X		X

-continued-

Homer creel survey schedule, 1989 (continued).

DATE	DAY	A (1000-1300)	B (1301-1600)	C (1601-1900)	D (1901-2200)
AUG 01	TUES	X			X
AUG 02	WEDS		X	X	
AUG 03	THUR	----------	OFF	----------	
AUG 04	FRI	----------	OFF	----------	
AUG 05	SAT		X		X
AUG 06	SUN		X	X	
AUG 07	MON	----------	OFF	----------	
AUG 08	TUES	----------	OFF	----------	
AUG 09	WEDS		X	X	
AUG 10	THUR		X	X	
AUG 11	FRI			X	X
AUG 12	SAT	X			X
AUG 13	SUN			X	X
AUG 14	MON	----------	OFF	----------	
AUG 15	TUES	----------	OFF	----------	
AUG 16	WEDS		X	X	
AUG 17	THUR			X	X
AUG 18	FRI		X	X	
AUG 19	SAT		X	X	
AUG 20	SUN		X	X	
AUG 21	MON	X			X
AUG 22	TUES	X			X
AUG 23	WEDS	----------	OFF	----------	
AUG 24	THUR	----------	OFF	----------	
AUG 25	FRI	X			X
AUG 26	SAT			X	X
AUG 27	SUN	X		X	
AUG 28	MON		X	X	
AUG 29	TUES	----------	OFF	----------	
AUG 30	WEDS	----------	OFF	----------	
AUG 31	THUR		X	X	
SEPT 01	FRI			X	X

Kodiak Creel Survey Schedule, 1989.

DATE	DAY	A (1000-1400)	B (1401-1700)	C (1701-2000)	D (2001-2300)
JULY 01	SAT		X		X
JULY 02	SUN	X	X		
JULY 03	MON		X	X	
JULY 04	TUES		X		X
JULY 05	WEDS	----------	DAY OFF	----------	
JULY 06	THURS	----------	DAY OFF	----------	
JULY 07	FRI	----------	DAY OFF	----------	
JULY 08	SAT		X	X	
JULY 09	SUN	X		X	
JULY 10	MON		X	X	
JULY 11	TUES			X	X
JULY 12	WEDS	X			X
JULY 13	THURS	----------	DAY OFF	----------	
JULY 14	FRI	----------	DAY OFF	----------	
JULY 15	SAT		X	X	
JULY 16	SUN	X		X	
JULY 17	MON	X			X
JULY 18	TUES		X	X	
JULY 19	WEDS	----------	DAY OFF	----------	
JULY 20	THURS	----------	DAY OFF	----------	
JULY 21	FRI		X	X	
JULY 22	SAT			X	X
JULY 23	SUN		X	X	
JULY 24	MON	X	X		
JULY 25	TUES			X	X
JULY 26	WEDS	X	X		
JULY 27	THURS	----------	DAY OFF	----------	
JULY 28	FRI	----------	DAY OFF	----------	
JULY 29	SAT	X		X	
JULY 30	SUN		X		X
JULY 31	MON		X	X	

-continued-

Kodiak Creel Survey Schedule, 1989 (continued).

DATE	DAY	A (1000-1400)	B (1401-1700)	C (1701-2000)	D (2001-2300)
AUG 01	TUES	X		X	
AUG 02	WEDS	X		X	
AUG 03	THUR	----------	DAY OFF	----------	
AUG 04	FRI	----------	DAY OFF	----------	
AUG 05	SAT	X	X		
AUG 06	SUN		X	X	
AUG 07	MON	X		X	
AUG 08	TUES		X	X	
AUG 09	WEDS	----------	DAY OFF	----------	
AUG 10	THUR	----------	DAY OFF	----------	
AUG 11	FRI		X	X	
AUG 12	SAT		X	X	
AUG 13	SUN	X			X
AUG 14	MON	X		X	
AUG 15	TUES		X		X
AUG 16	WEDS	X		X	
AUG 17	THUR	----------	DAY OFF	----------	
AUG 18	FRI	----------	DAY OFF	----------	
AUG 19	SAT		X	X	
AUG 20	SUN			X	X
AUG 21	MON		X		X
AUG 22	TUES	----------	DAY OFF	----------	
AUG 23	WEDS	----------	DAY OFF	----------	
AUG 24	THUR		X		X
AUG 25	FRI		X		X
AUG 26	SAT			X	X
AUG 27	SUN	X	X		
AUG 28	MON		X		X
AUG 29	TUES	----------	DAY OFF	----------	
AUG 30	WEDS	----------	DAY OFF	----------	
AUG 31	THUR		X	X	
SEPT 01	FRI			X	X
SEPT 02	SAT		X	X	
SEPT 03	SUN		X		X
SEPT 04	MON		X		X
SEPT 05	TUES	----------	DAY OFF	----------	
SEPT 06	WEDS	----------	DAY OFF	----------	
SEPT 07	THURS	----------	DAY OFF	----------	
SEPT 08	FRI		X	X	
SEPT 09	SAT	X		X	
SEPT 10	SUN	X		X	
SEPT 11	MON	----------	DAY OFF	----------	
SEPT 12	TUES	----------	DAY OFF	----------	
SEPT 13	WEDS	X	X		
SEPT 14	THURS			X	X
SEPT 15	FRI	X	X		

Sampling schedule for the Cordova Delta sport fishery, 1989.

Date	Strata[1]	Period/Time A:0600-0930	B:0931-1700	C:1701-2030
15-Jun	WD	X		X
16-Jun	WD	X		X
17-Jun	WE	X		X
18-Jun	WE		X	
19-Jun	WD		X	
20-Jun	WD			
21-Jun	WD			
22-Jun	WD	X		X
23-Jun	WD		X	
24-Jun	WE		X	
25-Jun	WE	X		X
26-Jun	WD			
27-Jun	WD			
28-Jun	WD	X		X
29-Jun	WD	X		X
30-Jun	WD	X		X
01-Jul	WE	X		X
02-Jul	WE		X	
03-Jul	WE	X		X
04-Jul	WE		X	
05-Jul	WD	X		X
06-Jul	WD			
07-Jul	WD			
08-Jul	WE	X		X
09-Jul	WE	X		X
10-Jul	WD	X		X
11-Jul	WD	X		X
12-Jul	WD			
13-Jul	WD			
14-Jul	WD	X		X
15-Jul	WE	X		X
16-Jul	WE	X		X
17-Jul	WD			
18-Jul	WD			
19-Jul	WD	X		X
20-Jul	WD	X		X
21-Jul	WD	X		X
22-Jul	WE		X	
23-Jul	WE	X		X
24-Jul	WD	X		X
25-Jul	WD	X		X

-Continued-

Sampling schedule for the Cordova Delta sport fishery, 1989 (continued).

Date	Strata[1]	Period/Time A:0600-0930	B:0931-1700	C:1701-2030
26-Jul	WD			
27-Jul	WD			
28-Jul	WD	X		X
29-Jul	WE	X		X
30-Jul	WE	X		X
31-Jul	WD	X		X
01-Aug	WD	X		X
02-Aug	WD			
03-Aug	WD			
04-Aug	WD	X		X
05-Aug	WE	X		X
06-Aug	WE	X		X
07-Aug	WD			
08-Aug	WD			
09-Aug	WD		X	
10-Aug	WD		X	
11-Aug	WD	X		X
12-Aug	WE	X		X
13-Aug	WE	X		X
14-Aug	WD			
15-Aug	WD			
16-Aug	WD	X		X
17-Aug	WD	X		X
18-Aug	WD		X	
19-Aug	WE	X		X
20-Aug	WE	X		X
21-Aug	WD		X	
22-Aug	WD			
23-Aug	WD			
24-Aug	WD	X		X
25-Aug	WD	X		X
26-Aug	WE	X		X
27-Aug	WE	X		X
28-Aug	WD			
29-Aug	WD			
30-Aug	WD	X		X
31-Aug	WD		X	
01-Sep	WD		X	
02-Sep	WE	X		X
03-Sep	WE	X		X
04-Sep	WE		X	

-Continued-

Sampling schedule for the Cordova Delta sport fishery, 1989 (continued).

Date	Strata[1]	Period/Time A:0600-0930	B:0931-1700	C:1701-2030
05-Sep	WD	X		X
06-Sep	WD	X		X
07-Sep	WD			
08-Sep	WD			
09-Sep	WE		X	
10-Sep	WE	X		X
11-Sep	WD	X		X
12-Sep	WD	X		X
13-Sep	WD			
14-Sep	WD			
15-Sep	WD	X		X
16-Sep	WE	X		X
17-Sep	WE		X	
18-Sep	WD	X		X
19-Sep	WD			
20-Sep	WD			
21-Sep	WD		X	
22-Sep	WD	X		X
23-Sep	WE		X	
24-Sep	WE		X	
25-Sep	WD		X	
26-Sep	WD	X		X
27-Sep	WD	X		X
28-Sep	WD			
29-Sep	WD			
30-Sep	WE	X		X
01-Oct	WE	X		X

[1] WD=Weekday, WE=Weekend/Holiday.

Sampling schedule for the Eyak sport fishery, 1989.

Date	Strata[1]	Period/Time A:0600-0930	B:0931-1700	C:1701-2030
15-Jun	WD	X		X
16-Jun	WD	X		X
17-Jun	WE	X		X
18-Jun	WE	X		X
19-Jun	WD			
20-Jun	WD			
21-Jun	WD	X		X
22-Jun	WD	X		X
23-Jun	WD		X	
24-Jun	WE	X		X
25-Jun	WE		X	
26-Jun	WD			
27-Jun	WD			
28-Jun	WD		X	
29-Jun	WD	X		X
30-Jun	WD	X		X
01-Jul	WE	X		X
02-Jul	WE	X		X
03-Jul	WE	X		X
04-Jul	WE	X		X
05-Jul	WD		X	
06-Jul	WD			
07-Jul	WD			
08-Jul	WE		X	
09-Jul	WE	X		X
10-Jul	WD			
11-Jul	WD			
12-Jul	WD	X		X
13-Jul	WD	X		X
14-Jul	WD	X		X
15-Jul	WE		X	
16-Jul	WE	X		X
17-Jul	WD		X	
18-Jul	WD	X		X
19-Jul	WD			
20-Jul	WD			
21-Jul	WD	X		X
22-Jul	WE	X		X
23-Jul	WE		X	
24-Jul	WD			
25-Jul	WD			

-Continued-

Sampling schedule for the Eyak sport fishery, 1989 (continued).

Date	Strata[1]	Period/Time A:0600-0930	B:0931-1700	C:1701-2030
26-Jul	WD		X	
27-Jul	WD		X	
28-Jul	WD	X		X
29-Jul	WE	X		X
30-Jul	WE	X		X
31-Jul	WD	X		X
01-Aug	WD	X		X
02-Aug	WD	X		X
03-Aug	WD			
04-Aug	WD			
05-Aug	WE		X	
06-Aug	WE	X		X
07-Aug	WD	X		X
08-Aug	WD			
09-Aug	WD			
10-Aug	WD	X		X
11-Aug	WD		X	
12-Aug	WE	X		X
13-Aug	WE	X		X
14-Aug	WD			
15-Aug	WD			
16-Aug	WD	X		X
17-Aug	WD	X		X
18-Aug	WD	X		X
19-Aug	WE	X		X
20-Aug	WE	X		X
21-Aug	WD	X		X
22-Aug	WD			
23-Aug	WD			
24-Aug	WD	X		X
25-Aug	WD		X	
26-Aug	WE	X		X
27-Aug	WE		X	
28-Aug	WD	X		X
29-Aug	WD	X		X
30-Aug	WD	X		X
31-Aug	WD			
01-Sep	WD			
02-Sep	WE	X		X
03-Sep	WE	X		X
04-Sep	WE		X	

-Continued-

Sampling schedule for the Eyak sport fishery, 1989 (continued).

Date	Strata[1]	Period/Time A:0600-0930	B:0931-1700	C:1701-2030
05-Sep	WD	X		X
06-Sep	WD	X		X
07-Sep	WD			
08-Sep	WD			
09-Sep	WE	X		X
10-Sep	WE	X		X
11-Sep	WD	X		X
12-Sep	WD	X		X
13-Sep	WD	X		X
14-Sep	WD			
15-Sep	WD			
16-Sep	WE		X	
17-Sep	WE		X	
18-Sep	WD	X		X
19-Sep	WD		X	
20-Sep	WD	X		X
21-Sep	WD			
22-Sep	WD			
23-Sep	WE		X	
24-Sep	WE	X		X
25-Sep	WD	X		X
26-Sep	WD			
27-Sep	WD			
28-Sep	WD	X		X
29-Sep	WD	X		X
30-Sep	WE	X		X
01-Oct	WE	X		X

1 WD=Weekday, WE=Weekend/Holiday.

Sampling schedule for the Eshamy sport fishery, 1989.

Date	Strata[1]	Period/Time[2] A:0800-1130	B:1131-1500	C:1501-1830	D:1831-2200
01-Jul	WE		H		
02-Jul	WE		H		
03-Jul	WE		H		
04-Jul	WE		H		
05-Jul	WD				L
06-Jul	WD				
07-Jul	WD				
08-Jul	WE	L			
09-Jul	WE			L	
10-Jul	WD				H
11-Jul	WD				L
12-Jul	WD				
13-Jul	WD				
14-Jul	WD		H		
15-Jul	WE		H		
16-Jul	WE			L	
17-Jul	WD				
18-Jul	WD				
19-Jul	WD		L		
20-Jul	WD	L			
21-Jul	WD			H	
22-Jul	WE			H	
23-Jul	WE			H	
24-Jul	WD		H		
25-Jul	WD			H	
26-Jul	WD	H			H
27-Jul	WD				
28-Jul	WD				
29-Jul	WE			L	
30-Jul	WE		H		
31-Jul	WD				
01-Aug	WD				
02-Aug	WD		H		
03-Aug	WD				L
04-Aug	WD			H	
05-Aug	WE			H	
06-Aug	WE			H	
07-Aug	WD			H	
08-Aug	WD				L
09-Aug	WD			H	
10-Aug	WD				

-Continued-

Sampling schedule for the Eshamy boat sport fishery, 1989 (continued).

Date	Strata[1]	Period/Time[2] A:0800-1130	B:1131-1500	C:1501-1830	D:1831-2200
11-Aug	WD				
12-Aug	WE	H			
13-Aug	WE		H		
14-Aug	WD		H		
15-Aug	WD			L	
16-Aug	WD		H		
17-Aug	WD				
18-Aug	WD				
19-Aug	WE			H	
20-Aug	WE			H	
21-Aug	WD			H	
22-Aug	WD			H	
23-Aug	WD				
24-Aug	WD				
25-Aug	WD				H
26-Aug	WE				H
27-Aug	WE		L		
28-Aug	WD			L	
29-Aug	WD		H		
30-Aug	WD				
31-Aug	WD				
01-Sep	WD			L	
02-Sep	WE		H		
03-Sep	WE		H		
04-Sep	WE			H	

1 WD=Weekday, WE=Weekend/Holiday.
2 H=High Use-stage Stratum, L=Low Use-stage Stratum.

APPENDIX

B. Instructions for Completing the Data Forms

Instructions for Completing the Marine Interview Forms.

It is important that the following information is included on the Angler Interview form for all locations:

Year; Month; Day; Survey Area; Site; Period (in the Harbor box); Counted; Continue (to denote that this row of species apply to the previous interview line); Class (type of boat); Target (specie); Area (Fished); Variables 1-2 (1-guided/unguided, 2-residency), Hours Fished (angler hours for freshwater surveys and angler days for harbor surveys).

Freshwater creel surveys will also record complete/incomplete. At Seward, variable field 4 will be used to record days fished.

Refer below and to the Mark-Sense Introduction Manual Developed by RTS for specific information.

Write in the date and page number on each sheet following the first sheet in a series. This information will be important in the event the pages are accidently mixed or out of order. Use a new sheet at the start of each period

Prince William Sound Creel Survey Codes:

Survey Site	Area	Site
Valdez Boat	J	001
Whittier Boat Passage Canal	J	002
Cordova Boat	J	728
Seward Boat Harbor	P1	002
Homer Boat Harbor	P1	003
Kodiak Boat Harbors	Q	745
Eyak River	J	005
Copper River Delta Beach		
Alaganik Slough	J	114
Clear Creek	J	133
Eshamy Creek and Lagoon	J	006
Anchorage PWS Charter	J	004

Harbor: To designate sample period.

Period	Selection
A	1
B	2
C	3
D	4

Class: Type of Boat

1: open boat
2: cabin cruiser (< 30')
3: cabin cruiser (≥ 30')
4: inflatable
5: kayak
6: sailboat

Target (Species)- record what the angler spent most of her/his time fishing for (can only mark one box!)
0: Coho (not salmonids as noted in the RTS Manual)
7: Sockeye (not bottomfish as noted in the RTS Manual)
A: Pink
B: Chinook
C: Halibut
D: Rockfish

Area Fished: - record where the angler spent most of his/her time (refer to maps for each area) (can only mark one box!)

Valdez, Cordova, Whittier
1: Valdez Arm
2: Western PWS Marine
3: Eastern PWS Marine
4: Island Freshwater

Seward
1. Resurrection Bay
2. Outer Coast - Western Bays
3. Outer Coast - Eastern Bays
4. Outer Islands

Homer
1. Kachemak Bay
2. Outside, N. of Anchor Pt.
3. Outside, N. of Pt. Pogibshi
4. Outside other

Kodiak
1. SE Afognak
2. Whale Island
3. Buoy 4
4. Inner Chiniak
5. Cape Chiniak

Variables 1-5:

Variable Field 1.
1= Unguided
2= Guided

Variable Field 2.
1= Local Alaska Resident (defined below)
2= Non-local Alaska Resident
3= Non Alaska Resident

Local:

Valdez:	a resident of Valdez
Cordova:	a resident of the Eastern PWS area (see map)
Whittier:	a resident of Whittier (don't include people who have recreational condos or boat slips in Whittier but live elsewhere most of the time)
Homer:	a resident of the Kenai Peninsula, south of Ninilchik (including Seldovia, Port Graham, English Bay)
Kodiak:	a resident of Kodiak or Afognak islands

Variable Field 4.
In Seward this field will be used to record Days Fished

Hours Fished: Days fished for boat surveys and angler hours for beach surveys. A two digit additive field, however you can not code a number higher than 9 in either column.

Species: Mark a species box.

Chinook salmon -	KS
Coho salmon -	SS
Pink salmon -	PS
Chum salmon -	CS
Sockeye salmon -	RS
Pacific halibut -	HA
Rockfish -	RF
Lingcod -	1
Dolly Varden char -	2
Cutthroat trout -	3
Cod, pollock -	4
Sculpin -	5
Skate -	6
Octopus -	7
Flounder -	8

Instructions for completing the Standard age weight length mark sense form

The following fields must be recorded:

Date (year, month, and day with leading zeros for month and day)

Location - where you are collecting the fish:

Survey Site	line 1	Line 2
Valdez Boat	10	001
Whittier Boat Passage Canal	10	002
Cordova Boat	10	728
Seward Boat Harbor	161	002
Homer Boat Harbor	161	003
Kodiak Boat Harbors	17	745
Eyak River	10	005
Copper River Delta Beach		
Alaganik Slough	10	114
Clear Creek	10	133
Eshamy Creek and Lagoon	10	006
Anchorage PWS Charter	10	004

Species:
200 = Halibut
130 = Lingcod
140 = Rockfish (will be further differentiated using the maturity index field)

Type of measurement:
Length: Total length = TL
Fishery: Sport Fishery = SPF

Page Number - Start over at 1 for each day. Be careful to enter this correctly if you are using the copy header option.

Sex

Maturity Index - used to designate maturity in rockfish
1 = eggs
2 = eyed larvae
3 = well developed eyed larvae

Length - record total length in millimeters (length from tip of tail to tip of snout)

Weight - used as a variable field to designate species of rockfish the numbers correspond to page numbers in Kramer & O'Connell's manual.

37 = quillback
11 = shortraker
61 = yelloweye
39 = black
17 = dusky
13 = silvergray
1 = rougheye
3 = Pacific ocean perch
5 = brown
7 = Aurora
9 = redbanded
15 = copper
19 = dark blotched
21 = splitnose
23 = greenstriped
25 = Puget Sound
27 = widow
29 = yellowtail
31 = chilipepper
33 = rosethorn
35 = shortbelly
41 = blackgill
43 = vermilion
45 = blue
47 = china
49 = tiger
51 = bocaccio
53 = canary
55 = northern
57 = redstripe
59 = yellowmouth
63 = stripetail
65 = harlequin
67 = pygmy
69 = sharpchin
71 = shortspine thornyhead
73 = longspine thornyhead

Instructions for completing the tag recovery form.

This form is to be used in a creel survey or field office when individual tags are returned that were each collected (caught) at a different location. Use of AWL forms is impractical in this situation because each new location would require a new AWL form. Do not use this form if you are collecting tags all from the same location (for instance at a weir); use an AWL form instead.

Circle the appropriate recovery type at the top of the page and write the location (such as Valdez, Cordova) below the recovery type. Number the pages consecutively for the entire year.

The Tag color codes are written at the bottom of the sheet - choose the appropriate code. Note: in Kodiak, pink tags in the range 5001-6000 must be carefully examined for a black stripe. Pink with a black stripe is WRBK.

The Recovery date is the date the fish was caught. If the angler can't recall the exact date, enter an approximate one.

The Area is the location where the fish was caught. Write in as exact a description as you can. Try to be consistent in your choice of place names for each site.

Site code - to be completed later by someone in RTS.

Fate - killed or released. If the fish is released, note in the comment field if the angler removed the tag.

Length (mm) - only enter a length here if you measure the fish. Angler's estimates of length or their length measurements can be written in the comment field. Measure the fish from tip of snout to fork of tail.

Sex - enter M or F.

Otolith - if you collect otoliths, enter Y, write the date, page, and line number on the otolith envelope.

Age - to be entered when the fish is aged.

APPENDIX

C. Instructions for Completing the Air Taxi Logbook.

Instructions for completing the air taxi logbook.

A logbook containing entries for the following questions will be distributed to all major air taxi operators that charter to PWS:

1. number of anglers in the party,
2. areas fished in PWS and points of drop-off,
3. number of hours fished by the party (by area),
4. target species (by area),
5. number of fish caught and harvested (by species and area),

Data from these questions will be kept on a log form to be distributed to the air taxis. The form design will allow for easy keypunching of the data.

APPENDIX

D. Chain of Custody Procedures

A "chain of custody" refers to the paper documentation that accompanies a sample, and which shows that the sample is under observation or control. Chain of custody procedures are invoked when there is a possibility that the study and its methods will be subjected to a legal challenge. The procedure ensures the integrity of each sample from time of collection until the analytical data are released. Documentation should occur at the following points in the process: 1) sample collection and preservation; 2) transportation to storage; 3) storage; 4) subsequent transportation for analysis; 5) acceptance for analysis; 6) analysis; 7) final disposition of samples. Each time the sample (or groups of samples) change hands, a record of the transfer is kept on a "Chain of Custody Transmittal Form," which indicates who has custody of the samples at all times.

SAMPLE COLLECTION

Information describing the collection of each sample is to be recorded in a field notebook and on a label permanently affixed to each sample jar (or written on each bulk sample on masking tape securing an aluminum foil-wrapped sample) to include: site description (location name, number, or other unique characteristics); date and local time; sampling effort name (vessel or team); and collector's name. The notebook should also include other observations (presence of oil, details for reoccupying the exact sample site, meteorological, hydrographical, and oceanographic conditions) and types of sampling used (hand, mechanical grab, trawl, beach seine), personnel involved (name, address, telephone number, and affiliation), or other pertinent remarks. A unique sample number can also be assigned, preferable as a alphanumeric to reduce potential overlap of numbers between different groups.

When sampling is completed at each site, a Chain of Custody Transmittal Form is initiated for each different type of sample (organisms by species for chemistry, sediments for chemistry, organisms for taxonomy or other purposes, water samples, bacteriology samples, etc.).

Each project team will maintain its own sequence of sample numbers as follows:

Project Number	Agency	Field Team	Date	Daily Sequence
00	"A"	"a"	000000	0000

This information will be included on the sample label and the "Chain of Custody Transmittal Form." Also enter "Action Taken," e.g., "sample collection" on the form. Both the collector and a witness must sign the form for each set of samples. The Chain of Custody Transmittal Form is then kept in close physical proximity to the samples; if the samples are stored in a freezer or on ice, the sheets may be kept in the field notebooks. Not only does this form establish the existence of the samples, but it also provides an initial inventory of the samples collected.

TRANSPORTATION OF SAMPLES FOR STORAGE

If the samples are periodically removed from the field and transported to a central storage facility under control of anyone except the primary collector, add another line on the Chain of Custody Transmittal Form with the date and time that the samples were transferred from the primary collector to the person responsible for the transportation, total number of samples, action taken and signatures.

If a collection, or sample is to be separated so that some individuals will be preserved for hydrocarbon analysis and others for histological preparation, separate Chain of Custody Transmittal Forms must accompany each part. It may be possible to simply photocopy the form at time of sample splitting in such cases.

Appendix

E. FY90 Operational Plan:

Sport Harvest with Estimates of Stocked Contribution for Resurrection Bay Fisheries

This plan was not prepared as part of the oil spill assessment planning process. Rather, it was prepared as an internal Alaska Department of Fish and Game, Sport Fish Division, plan to guide ongoing evaluations of stocking programs being conducted in the Resurrection Bay drainage. Some of aspects of this plan may have been changed in-season to accomdate oil spill assessment work. These changes are not reflected in this document.

CONFIDENTIAL

DRAFT

STATE/FEDERAL NATURAL RESOURCE DAMAGE ASSESSMENT
DETAILED STUDY PLAN

Project Title:	INJURY TO PINK/CHUM SALMON SPAWNING AREAS OUTSIDE PRINCE WILLIAM SOUND
Study ID Number:	Fish/Shellfish Study Number 7
Lead Agency:	State of Alaska, ADF&G; Commercial Fisheries Division
Cooperating Agency(ies):	Federal: USFWS State : None
Principal Investigators:	Charles Swanton, Fishery Biologist Henry Yuen, Fishery Biologist
Assisting Personnel:	Jeff Fox, Fishery Biologist Kevin Brennan, Fishery Biologist
Date Submitted:	October 9, 1989

	Signature	Date
Principal Investigators:		10/10/89
	Henry Yuen	10/11/89
Supervisor:		
OSIAR Senior Biometrician:		
OSIAR Program Manager:		
OSIAR Director:		

I. OVERVIEW

This study plan contains two separate sections. These sections address the general objectives outlined in Fish Study Number 7 for Lower Cook Inlet/Kenai Fjords and the Kodiak and Chignik Management areas, respectively. These areas had major differences in oil distribution, and they also differ in the relative importance of intertidal spawning, and the magnitude of the run returns in 1989. In the Cook Inlet/Kenai Fjords area, escapement of pink and chum salmon was generally modest, whereas streams in the Kodiak/Chignik area had some very large escapements because of almost complete closure of the commercial seine fishery. On the other hand, oil directly impacted intertidal spawning areas more in the Cook Inlet/Kenai Fjords area than in the Kodiak/Chignik area and in the Kodiak/Chignik area there are fewer intertidal spawning areas for pink and chum salmon than in Cook Inlet and the Kenai Fjords. Because of these differences, the damage assessment studies in each area were designed differently. In the Cook Inlet/Kenai Fjords area the study is designed to determine the direct impacts of oil on survival of pink and chum salmon in intertidal spawning areas. In the Kodiak/Chignik area, the study is designed to evaluate an indirect effect of the oil spill (overescapement) on pink and chum salmon production. In addition, Fish Study Number 8 is an interrelated component of this study and is also designed to assess the damage of the oil spill on pink and chum salmon.

II. LOWER COOK INLET/KENAI FJORDS STUDIES

INTRODUCTION

This project was designed to evaluate the distribution of pink and chum salmon spawning in intertidal and upstream areas as a result of oil contamination from the Exxon Valdez oil spill. This project also provides spawner distribution data for the pink and chum salmon egg and pre-emergent fry study (Fish Study No. 8). Intertidal spawning areas are important to pink and chum salmon, and these areas are much more vulnerable to contamination by an oil spill than upstream spawning areas.

The Alaska Department of Fish & Game Lower Cook Inlet management area (LCI) pink and chum salmon spawning ground escapement survey project is an ongoing study that provides data on numbers of live salmon by species for 27 streams at 14 day intervals. This oil spill assessment project will expand upon the existing project by increasing survey frequency, resuming carcass counts, and mapping spawner distribution within the intertidal areas for eight of those streams.

OBJECTIVES

1. Estimate numbers of salmon by species within standardized intertidal and upstream zones for eight streams in the Lower Cook Inlet/Kenai Fjords area.

2. Produce a catalog of aerial photographs and/or detailed maps of spawner distribution for the more important pink and chum salmon streams in the Lower Cook Inlet/Kenai Fjords area for use in designing sampling transects in the egg and pre-emergent fry studies (Fish Study No. 8).

3. Determine the presence or absence of oil on intertidal habitat used by spawning salmon through visual observation and hydrocarbon analysis of tissue samples from intertidal mussels (Mytilus sp.) collected in the immediate area of each sampled stream.

4. Identify potential alternative methods and strategies for restoration of lost use, populations, or habitat where injury is identified.

METHODS/DATA ANALYSIS

Study Design

This project is designed to evaluate changes in numbers and distribution of spawning salmon relative to oil contamination from the Exxon Valdez spill of March 1989. Two crews of two people each will perform foot surveys of intertidal and upstream portions of eight major pink and chum salmon spawning streams. Each stream will be surveyed at least once a week during low tide. The spawning season is approximately between July 7 and September 7.

Streams to be surveyed will be selected using the following criteria:

1. The stream is included in the aerial survey program.
2. The stream was examined in past spawning ground survey programs.

During each stream survey the following data will be recorded (Appendix A):

1. Stream name;
2. Date and time;
3. Counts of live and dead salmon by species and location in the stream (0.0-0.6 m, 0.6-1.2 m, and 1.2-1.8 m below mean high water and above tidal inundation);
4. Comments on the quality of the survey: visibility, completeness of survey, etc.
5. Observers names.

A map of the intertidal zones will be drawn for each stream. Landmarks for the boundaries between intertidal zones, distances across the streams with zones, distances between zone boundaries, areas of spawning concentration, and areas of preference by species within each intertidal zone will be recorded on the maps. These maps will be used later by the Pink and Chum Salmon Egg and Pre-emergent Fry Sampling project (Fish Study No. 8).

Early in the season, one of the crews will mark the tide levels at each stream. Intertidal zones in the Lower Cook Inlet/Kenai Fjords area will be measured from the mean high tide level due to large differences in mean tidal height between the Gulf of Alaska (4 m) and Cook Inlet (6 m) sides of the Kenai Peninsula. The stream bed location of tide levels 0.0, 0.6, 1.2, and 1.8 m below mean high water will be marked with a 0.3 m^2 fluorescent orange plywood rectangle. The markers will be numbered consecutively 1 through 4 with number 1 furthest downstream at the 1.8 m below mean high water level. A commercial hand held tide computer with specific time and location corrections will be used to determine tide heights. Stream length within a zone will be measured as well as width at 25 m intervals.

A composite sample of mussels (Mytilus sp.) will be collected at

the mouth of each stream for hydrocarbon analysis. A field blank (sample container opened at the collection site, closed and stored as if it contained a sample) and two sample replicates will also be collected. Results of the analysis will be used to document the level of oil impact sustained by the stream. Each sample will consist of enough mussels to provide 10 grams of tissue for analysis. The mussels will be collected from the immediate vicinity of all streams. Collectors will use wooden tongue depressors when possible and clean hands otherwise. All mussels will be above water when collected to prevent contamination by surface hydrocarbons. The sample containers will be pre-rinsed (with dicloromethane) glass jars with teflon lined lids as supplied by I-Chem. The samples will be stored in padlocked containers and kept in a freezer in the Homer ADF&G office. Appropriate chain of custody forms will accompany each sample.

Both crews will count the numbers of live and dead salmon within each of the four tide zones (0.0-0.6 m, 0.6-1.2 m, and 1.6-1.8 m below mean high tide, and upstream of tidal inundation) for all eight streams. Surveys always begin at the 1.8 m below high tide line and progress upstream. Hence, all surveys will be start at low tide. Both live and carcass counts will be made while walking upstream for a tide zone before continuing on to the next zone. The upstream limit of a survey will be determined by the presence of natural barriers to fish passage (i.e. waterfalls), by the end of the stream, or by the absence of spawning salmon. Hand tally counters will be used when counting. Both crew members will walk the intertidal and single channel stream areas together but count independently. Both crew members will periodically compare their counts. They will recount as many times as necessary until their count differ by 10% or less. The average of each crew member's counts will be recorded unless one member feels that his/her count was invalid. Where the stream forks, the crew will split and walk the remaining areas alone. Survey partners will be rotated on a regular basis to minimize counting biases.

Study Sites

The eight streams selected for this project are Windy Creek Left, Port Dick Creek, Windy Creek Right, and Island Creek in the Kenai Fjords area and Humpy Creek, China Poot Creek, Seldovia River, and Port Graham Creek in the Cook Inlet area. The first two creeks have had oil deposited near the stream mouths, the next two have had oil floating offshore, and the remainder may have had no impact.

Data Analysis

Total escapement to each stream will be estimated using counts from the foot surveys and stream life estimates. Estimation methods will be similar to those used by Johnson and Barrett (1988) and

stream life will be from historical data or estimated from data collected in this study.

Streams will be divided into 2-3 categories based on levels of hydrocarbon contamination (as determined from visual observations and hydrocarbon level in mussel tissues). Counts of salmon by species and stream zone for each stream will be assigned to one of the hydrocarbon categories. Categorical data analysis techniques such as log linear models using chi-square statistics will be used to look for differences in numbers of spawning salmon by stream zone and stream and relate any differences to the level of hydrocarbon contamination. Counts and spawner distribution data will also be compared with historical stream survey data and related to the level of hydrocarbon impact.

Statistics that will be estimated include:

1. Counts of spawning and dead salmon by stream zone, stream, and date will be used to in the comparison of spawner density with egg and fry density.
2. Stream life in order to estimate total spawning escapement by species for each stream.

SCHEDULES AND REPORTS

Date(s)	Activity
Jul 7 - Sep 7 1989	Replicate surveys of 8 streams.
Sep 8 - Oct 20 1989	Research historical data, data entry, analysis, and report.

PROJECT BUDGET [1]

Line Item	Category	Budget
100	Personnel Services	$ 15,500
200	Travel	$ 1,800
300	Contractual	$ 15,000
400	Commodities	$ 2,600
500	Equipment	$ 6,200
700	Grants	$ 0
Total		$ 41,100

[1] Budget is for all activities performed from March 27, 1989 to February 28, 1990.

FUNDED PERSONNEL

Class	PCN	Name	PFT_mm	SFT_mm
FB II	11-1258		2.0 (funded under CF-381)	
FB I	new			1.5
FT II	11-1536			1.0
FT II	11-1516		(funded under CF-383)	1.0

LITERATURE CITED

Johnson, B.A. and B. Barrett. 1988. Estimation of salmon escapement based on stream survey data. Alaska Department of Fish and Game. Division of Commercial Fisheries. Regional Information Report No. 4K88-35. Kodiak.

III. KODIAK AND CHIGNIK MANAGEMENT AREA STUDIES

INTRODUCTION

The intent of this investigation is to provide area wide estimates of pink and chum salmon escapement for the Kodiak and Chignik Management areas (Figure 1). During 1989 the presence, close proximity, or threat of oil contamination from the EXXON VALDEZ oil spill, prohibited the commercial harvest of pink and chum salmon which were in excess of target escapement goals for these areas. Harvest derived from these species is an important component (1969-1987 odd year averages of 87% = 6,517,000 pinks and 745,000 chum for Kodiak; 35% = 705,097 pinks and 165,725 chum for Chignik) of the total annual salmon catch (Malloy 1989; Thompson and Fox 1989). Ex-vessel value of the combined pink and chum salmon catch has averaged 14.8 and 3.8 million dollars respectively, for Kodiak and Chignik over the last 9, odd years (Malloy 1989; Thompson and Fox 1989).

Aerial stream surveys, using fixed wing aircraft and trained observers, have been conducted annually since the 1940's on selected streams in the Kodiak and Chignik Management areas (Larry Malloy ADF&G, Kodiak, personal communication). This information is used for estimating escapements and forecasting runs. Although counts were not made every year, these data provide an index for assessing brood year escapement trends and returns. Weirs on stream systems in the Kodiak and Chignik Management areas have a long history of use. Data collected from weir counts and samples permit assessment of total escapement and brood year returns of salmon species for these systems.

Estimates of total escapement of pink and chum salmon by stream for the Kodiak and Chignik areas will enable the calculation of foregone harvest and revenues for 1989, and provide a foundation for investigations responsible for quantifying effects of excessive escapements upon future brood year production (Fish Study No. 8).

OBJECTIVES

A. Estimate the numbers of spawning salmon by species and by intertidal and upstream areas for pink and chum salmon streams outside the sound where historical fry density data exist. These include 43+ streams in the Kodiak Island/Shelikof Strait Mainland area and 18 streams in the Chignik area. This objective entails:

   1. Using aerial survey data, to develop escapement indices for streams where historical fry density data exist.

   2. Ascertaining average pink salmon stream life using weir, aerial and foot survey counts of live and dead fish in selected streams.

3. Deriving total escapement using quantitative models by stream and management area which incorporate stream life and aerial survey escapement data.

B. Produce a catalog of aerial photographs and detailed maps of spawner distribution of pink salmon for index streams in the Kodiak and Chignik areas. These maps will allow for developing an enhanced pre-emergent fry sampling design for Fish Study Number 8.

C. Identify potential alternative methods and strategies for restoration of lost use, populations, or habitat where injury is identified.

METHODS

Two components comprise this study: 1) repeated aerial surveys for escapement indices on existing pre-emergent fry index streams; and 2) weir stations located on selected streams to count fish, collect carcasses, and perform foot surveys for enumerating live and dead pink salmon. State/Federal Natural Resource Damage Assessment Plan Fish/Shellfish Study numbers 7 and 8 are interrelated, therefore, methods and data collected in this study will be used and applied in Fish Study Plan 8.

Aerial Surveys

Trained fisheries biologists will conduct aerial surveys using fixed wing aircraft on the 43 Kodiak and 18 Chignik Management area pre-emergent fry index streams. Surveys will be flown weekly on each index stream with the program continuing until seven surveys over the spawning season (optimally including peak spawning activity) have been conducted or when spawner counts decrease to less than 10 percent of the maximum peak count observed. Additional nonindex streams (323 Kodiak and 72 Chignik) will be surveyed as time and aircraft availability permit. For each stream the following data will be recorded: 1) stream name and statistical number; 2) Julian date, weather conditions, visibility rating (bay, mouth and stream), and time survey conducted; 3) observer, pilot and aircraft type; 4) number of live and dead fish of each species (in bay, mouth and stream reach); and 5) general comments. Data will be recorded in field notebooks then transferred to survey form CFAS01 (Appendix A.1). Subsequently, survey data will be entered into the regional survey data base. The observer upon completion of the surveys for a particular index stream will map spawner distribution and designate numbers of live and dead fish by species on an enlarged U.S.G.S. topographical map. For a more in-depth discussion of aerial survey methods refer to Bevan (1961) or Cousens et al. (1982).

Weir Stations

Weirs will be located at two established sites (Akalura and Horse Marine Creeks) and two new sites (Afognak River tributary and East Arm Paramanoff Creek). For contrast of stream life information, two weir locations are on the south end of Kodiak Island (Akalura and Horse Marine) and two north are on the north on Afognak Island (Afognak River tributary and East Arm Paramanoff). Pink salmon weir counts will be made on a daily basis and sequestered from personnel responsible for pink salmon aerial survey counts in an effort to eliminate a priori knowledge of actual counts while aerial surveys are conducted. Daily and cumulative weir counts will be recorded on form CFWR-01 (Appendix A.2). Weir crews and additional personnel will do foot surveys of Pillar, Akalura, Horse Marine, East Arm Paramanof, and Afognak River tributary creeks for counts of live and dead pink salmon on a minimum of two and maximum of three day rotational basis. Surveys will be initiated prior to appreciable numbers of pink salmon entering streams and curtailed during periods of high flow or when turbidity impedes visibility. Live and dead count data will be recorded on form AKA89-1 (Appendix A.3). For all streams where periodic live and dead fish counts are conducted, a time series of up to seven weeks of counts could be available, providing a maximum of 21 data points per system. Data collected from weir operations will be used for developing average stream life, correction factors for aerial survey calibration, and expansion of aerial survey escapement indices to total escapement by stream and management area.

DATA ANALYSIS

Stream Life Calculation

Stream life for a pink salmon is defined as the length of time spent in a particular stream from initial fresh water entry extending to when the fish expires or exits the system. On a population level, average stream life can be determined using the difference between peak counts of live and dead fish in days, over time. For this study, stream life will be determined for each of the five streams where live and dead fish count data will be collected. In calculating average stream life, live and dead fish counts are assumed to be normal probability distributions (Mundy 1979; Eggers 1984). Stream specific live and dead count distributions will be evaluated to determine if this assumption is satisfied using graphical methods. To determine whether specific system stream life values can be combined, differences will be assessed by system from an average of all stream life values. If specific values are less than one standard deviation from the overall mean, the overall stream life value will be used for all forthcoming analyses. If differences exist, a stratification scheme will be imposed to determine which stream life values will be utilized for specific index stream escapement calculations. The stratification system, if required, will be founded on the

following variables; stream order, orientation, geomorphology, stream length and climatology.

Alternative determination of stream life values for each weired system will be formulated using a model derived by Johnson and Barrett (1988). Using known temporal escapement counts and cumulative total escapement, stream life will be estimated by setting an arbitrary stream life value and incrementing the value (increasing or decreasing) until the model converges upon the known cumulative total escapement. This process will be repeated for all four weir systems. Using this model provides an objective unbiased evaluation of stream life values derived from the difference in live and dead fish distribution method.

Total Escapement Estimation

Temporal aerial survey escapement counts of pink salmon in spawning streams are, depending upon the frequency and timing of the surveys, related to total or cumulative escapement. Defining and quantifying variables or relationships that allow transformation of aerial survey counts into reliable estimates of total escapement is the major question. Nelson and Geen (1981) derived a methodology for expanding aerial survey counts to total escapement based upon stream residence time of female chinook salmon. Bevan (1961) has identified and evaluated types of variability and bias in aerial survey methods. The contemporary premise is that given a series of aerial survey counts and some measure of stream life or stream residence time, a reasonable estimate of total escapement can be determined. Within the confines of this study, two approaches to estimating total escapement by stream and management area will be utilized.

The first approach is a model devised by Eggers (1984) which incorporates the parameters s - standard deviation of fish entry into a stream, SL - stream life and a - carcass wash out rate. The model, given values for the parameters, estimates an aerial survey correction factor which allows for conversion of peak aerial survey counts to total escapement by stream. All parameters can be derived from data collected at weir stations. Assumptions of the model are: 1) temporal entry of fish into a stream follows a normal probability distribution; 2) stream life is normally distributed; 3) number of dead fish removed from a stream in a given day is proportional to the number of dead salmon in the stream in that given day. The third assumption is the only one which has not been addressed to this point and will be evaluated using dead fish counts from stream surveys and carcass accumulations on weir panels.

A second model which can be invoked to address estimating total escapement from aerial survey counts is the geometric approach presented by Johnson and Barrett (1988). Two data requirements are necessary for the algorithm; escapement counts over time and stream life. The unit of measurement is area of the spawner abundance curve derived from the series of escapement counts. There are two

elements within the approach, first is calculating number of fish between survey counts and the second is calculating total escapement. A suite of scenarios or cases can be accounted for which promotes flexibility with regard to data requirements. Accuracy of the escapement estimate according to Johnson and Barrett (1988) is a function of the accuracy of the escapement counts and to a lesser extent the stream life estimate.

Both models will be assessed for reliability by using known counts applied to the models in addition to comparing the total escapement values by stream. Assessing the reasonableness of the escapement totals will provide a measure of the applicability of each model applicability.

Spawner Distribution Mapping and Aerial Photography

Spawner distribution maps will be prepared in part by observers conducting aerial escapement surveys and completed by personal responsible for surveys conducted in Fish Study Number 8. Aerial photography will be conducted in a similar fashion for each index stream surveyed.

SCHEDULES AND REPORTS

Dates	Function
June - September	Aerial and foot surveys, data collection
October - November	Data editing, analysis, report and map preparation
November 1	Preliminary report - total area wide escapement
November 2 - December	Data analysis, report writing
December 28	Total escapement - Final Report
December 29 - January 1990	Refinement and Evaluation, Map finalization

PROJECT BUDGET

Line Item	Category	Budget[1]
100	Personnel	$117,100
200	Travel	2,400
300	Contractual	132,200
400	Supplies	20,400
500	Equipment	7,100
700	Grants	-0-
Total		$279,200

[1]Budget encompasses activities performed from 15 July 1989 to 28 February 1990.

FUNDED PERSONNEL

Class	PCN	Name	MM	OT	SEA	HAZ	COST
FB IV	1202	D. Schmidt	0.0	0	0	0	-0
FB III	7016	B. Barrett	0.0	0	0	0	-0
FB II	7017	C. Swanton	6.8	350	0	0	38,595.00
FB II	1419	J. Fox	0.5	180	0	0	8,186.00
FB I	N315	A. Dill	3.0	240	0	0	14,818.00
FB I	1417	P.Roche	5.3	250	0	0	30,914.09
FT III	1596	P. Kuriscak	0.0	180	0	0	4,608.13
FT III	1597	D. Kaplan	3.0	240	0	0	17,631.08
FT III	1652	C. Neff	0.0	180	0	0	4,692.18
FT III	N415	S. Rodeo	2.3	180	0	0	9,233.84
FT III	N416	P. Ralston	1.5	120	0	0	6,155.89
FT III	N418	L. Brockman	1.5	120	0	0	6,155.89
FT II	1342	C. Hemke	2.0	128	0	0	10,475.06
FT II	N409	L. Ranallo	2.3	180	0	0	8,175.84
FT I	1472	R. Stoehr	0.0	80	0	0	1,527.15

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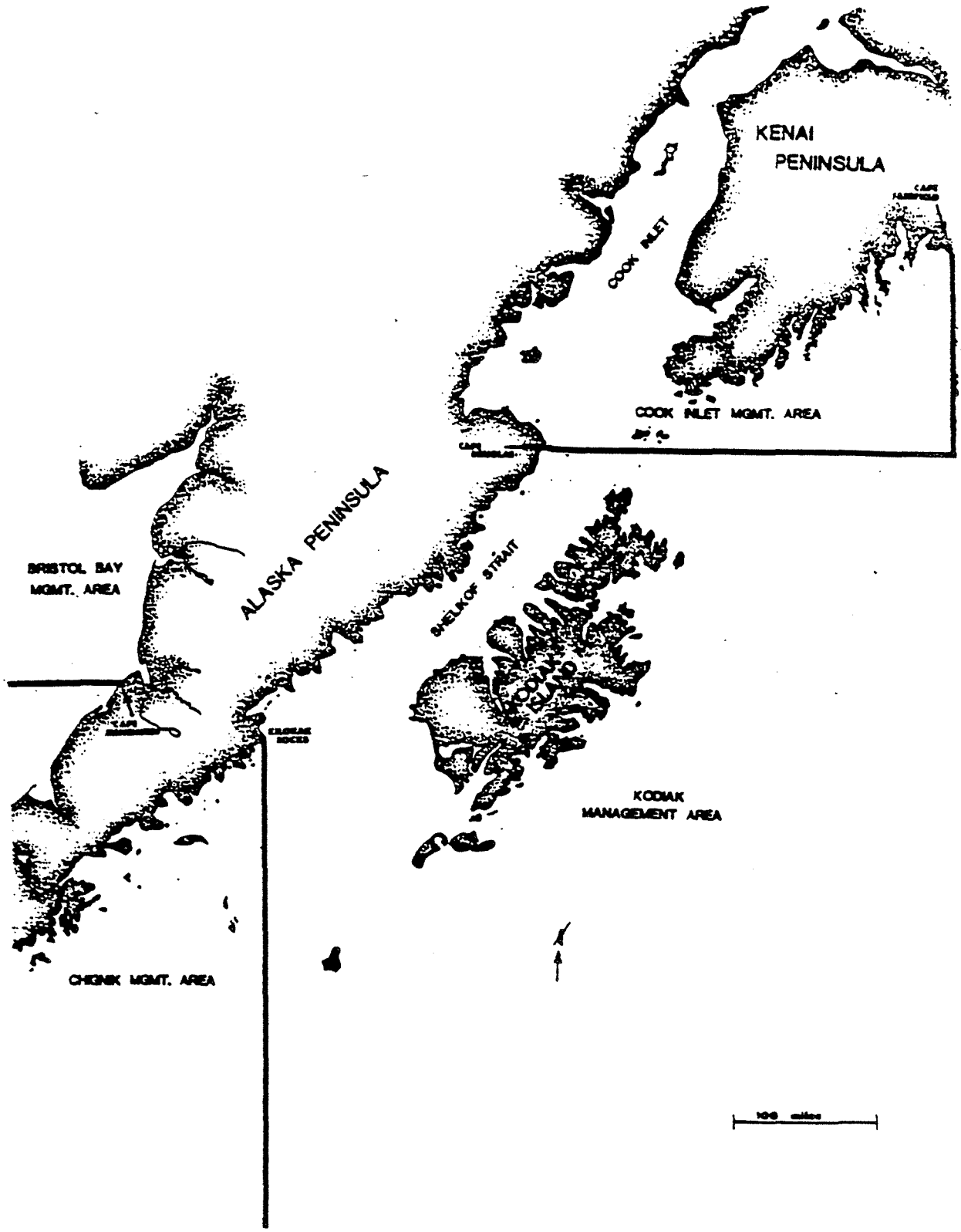

Figure 1. Map demarcating the Kodiak and Chignik Management Areas (Barrett and Monkiewicz 1989).

Appendix A.1. Aerial survey data recording form.

FORM: CFAS01

AERIAL SURVEY
KODIAK MANAGEMENT AREA
STREAM SURVEY LOG

OBSERVER: ____________________

PILOT ____________________

TIME IN: __________ TIME OUT: __________

AIRCRAFT: ____________________

WEATHER: ____________________

DATE: ____________________

TIDE: ____________________

Stream		Stream Visibility				Numbers[1] of Fish by Location				
No.	Name	P	F	G	E	Species	Bay	Mouth	Stream	Remarks
						Pink				
						Chum				
						Pink				
						Chum				
						Pink				
						Chum				
						Pink				
						Chum				
						Pink				
						Chum				
						Pink				
						Chum				
						Pink				
						Chum				

[1] Count reflects only live fish unless indicated in the "Remarks" column.

ALASKA DEPARTMENT OF FISH AND GAME
KODIAK MANAGEMENT AREA
WEEKLY SALMON WEIR CAMP REPORT FOR YEAR:_____

Form: CFWR-01

WEIR CAMP NAME:_____ PERSONNEL:_____

WEEKLY REPORT NO:_____ FOR WEEK ENDING (SATURDAY)_____

DATE		DAILY TOTAL SALMON ESCAPEMENT KINGS	REDS	COHO	PINK	CHUMS	DAILY TOTALS	JACK NO.	REDS %	STEELHEAD DOWN	STEELHEAD UP	DOLLYS UP	GILLNET MARKED REDS	GILLNET MARKED OTHER	H_2O LEVEL UP	H_2O LEVEL DN	H_2O TEMP.	WEATHER CEIL.	WEATHER VIS.	WEATHER WIND DIR./ SPEED
SUN	D																			
	A																			
MON	D																			
	A																			
TUE	D																			
	A																			
WED	D																			
	A																			
THU	D																			
	A																			
FRI	D																			
	A																			
SAT	D																			
	A																			
TOTAL FOR WK ____																				
ACCUM. THRU WK ____															COMMENTS					
WEIR MORTS. FOR WK															COMMENTS					
TOT. ACCUM.																				
AWL SAMPL. FOR WK													COMMENTS							
TOT. ACCUM.																				

ADDITIONAL COMMENTS: (BEAR AND PEOPLE PROBLEMS: SMOLT MIGRATION: WEIR PROBLEMS: CABIN REPAIR: NOTE AIRCRAFT TRAFFIC

Appendix A.2. Weekly weir report form.

Appendix A.3. Foot survey data recording form.

PINK SALMON STREAM SURVEY FORM

AKA89-

			STREAM						
			Survey		Counts			Temp.1/	
Stream Name	Date	Obser.	Cond.	Meth.	Live	Dead	Total	°C	Remarks

[1]Stream temperature should be recorded in celsius and taken at the approximate midpoint of the particular stream.

CONFIDENTIAL

STATE/FEDERAL NATURAL RESOURCE DAMAGE ASSESSMENT
DETAILED STUDY PLAN

Project Title: INJURY TO PINK AND CHUM SALMON EGG AND PRE-EMERGENT FRY OUTSIDE PRINCE WILLIAM SOUND

Study ID Number: Fish/Shellfish Study Number 8

Lead Agency: State of Alaska, ADF&G;
Commercial Fisheries Division

DRAFT

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

Principal Investigator: Charles Swanton, Fishery Biologist
Henry Yuen, Fishery Biologist

Assisting Personnel: Kevin Brennan, Fishery Biologist
Jeff Fox, Fishery Biologist

Date Submitted: October 9, 1989

	Signature	Date
Principal Investigator:		10/10/89
		10/11/89
Supervisor:		10/11/89
OSIAR Senior Biometrician:		
OSIAR Program Manager:		
OSIAR Director:		

I. OVERVIEW

This study is designed to determine the effects of the Exxon Valdez oil spill on abundance and survival of pink and chum salmon eggs and fry in the Lower Cook Inlet area (LCI) and Kodiak Island area. Because of different stream morphology, spawner distribution, variability in oiling, and differing rates of harvest of surplus salmon in the Kodiak and LCI areas, the study areas are addressed in separate sections of this study plan. These studies are closely tied with the studies of pink and chum salmon spawner distribution (Fish Study Number 7).

II. LOWER COOK INLET AND KENAI FJORDS AREA STUDIES

INTRODUCTION

This project was designed to evaluate pink and chum salmon egg to fry survival in the intertidal spawning areas affected by the Exxon Valdez oil spill. Intertidal spawning areas are important to pink and chum salmon in the LCI area and these areas are vulnerable to oil spills.

From 1964 through 1984, the Alaska Department of Fish and Game conducted pre-emergent fry digs annually on 24 streams in the Lower Cook Inlet/Kenai Fjords area. These fry density data were essential for forecasting future adult returns of pink salmon, however, funding for this study was cut from Fish and Game's general fund projects in 1985. This project will reinstate the pre-emergent fry study, provide additional information on egg density, and expand upon the level of sampling in eight of the 24 streams so that egg to fry survival can be estimated.

OBJECTIVES

1. Assess overwinter survival (eggs to pre-emergent fry) for pink and chum salmon eggs in oiled and non-oiled areas.

2. Assess loss in production, if any, from changes in overwinter survival.

3. Identify potential alternative methods and strategies for restoration of lost use, populations, or habitat where injury is identified.

METHODS/DATA ANALYSIS

Study Design

Sampling will be conducted in two phases: egg-digs which are performed in late October-early November, and pre-emergent fry digs

conducted in mid-March to mid-April.

Streams to be sampled were selected using the following criteria:

1. A sufficiently large adult salmon return to indicate a high probability of success when performing egg and fry digs.
2. Past history of egg or fry dig sampling.
3. Streams included in spawning ground surveys (Fish Study Number 7) and aerial escapement survey project (an existing Commercial Fish Division project).
4. Logistic ease and safely of sampling during the winter and early spring months.

Study Sites

The eight streams selected for this project are Windy Creek Left, Port Dick Creek, Windy Creek Right, and Island Creek in the Kenai Fjords area and Humpy Creek, China Poot Creek, Seldovia River, and Port Graham Creek in the Cook Inlet area. The first two creeks have had oil deposited near the stream mouths, the next two have had oil floating offshore, and the remainder may have had no impact from oil.

Sample Design

Sampling methods are identical for the pre-emergent fry and egg digs. On each sample stream, four zones, three intertidal and one above tidal inundation, will be identified and marked by crews conducting stream surveys during the Injury to Pink/Chum Salmon Spawning Areas project (Fish Study Number 7). The zones are 0.0-0.6 m, 0.6-1.2 m, and 1.2-1.8 m below mean high water, and upstream of tidal inundation.

Separate linear transects 30.5 m in length will be established in each zone (one transect for each type dig). The transects will run diagonally across the river with the downstream end located against one bank and the upstream end against the opposite bank. Overlapping of transects will be kept to a minimum to control the influence of fall egg digs on abundance of fry during spring sampling. Fourteen circular digs (56 per stream), each 0.3 m^2 in size, will be systematically dug along each transect using a high pressure hose to flush eggs and fry from the gravel. Eggs and fry will be caught in a specially designed net. Areas where salmon were not observed spawning during the spawning ground surveys (Fish Study Number 7) will be avoided. Numbers of live and dead fry by species as well as numbers of live and dead eggs by species will be collected from each 0.3 m^2 dig. Additional information such as date, time, and zone will also be collected.

Two samples of eggs and alevin will be collected from each of the 0.6-1.2 m below mean high water stream zones for hydrocarbon analysis. A field blank (sample container opened at the collection site, closed and stored as if it contained a sample) will also be

collected. Results of the analysis will be used to document the level of oil impact sustained by the stream, eggs, and alevins. Each sample will consist of enough eggs or fry to provide 10 grams of tissue for analysis. Collectors will avoid putting the samples into contact with any plastics, latex, etc. when gathering the samples. The sample containers will be pre-rinsed (with dicloromethane) glass jars with teflon lined lids as supplied by I-Chem. The samples will be stored in padlocked containers and kept in a freezer in the Homer ADF&G office. Appropriate chain of custody forms will accompany each sample.

Data Analysis

The major objective of this analysis is to test for differences in egg to fry survival between streams which were oiled and those that were not. The power of the test is unknown; consequently, the number of streams sampled is based on what can be surveyed in a reasonable manner given the window of time during which sampling must take place.

A mixed effects analysis of covariance will be used to test for differences in egg to fry survival due to oiling. Level of hydrocarbon impact and height in the tidal zone will be treated as fixed effects. Height in the tidal zone is nested within stream, a random effect. The level of hydrocarbon impact will be determined from hydrocarbon analysis of mussels collected by the Injury to Pink/Chum Salmon Spawning Areas project (Fish Study Number 7).

Analysis of variance will be used if no suitable hydrocarbon data are available. Degree of oiling as visually assessed by the Injury to Pink/Chum Salmon Spawning Areas (Fish Study Number 7) will be used to post-stratify streams. Degree of oiling will be treated as a fixed effect and height in the tide zone will also be a fixed effect nested in stream, a random effect.

An assessment of lost fry production will be made if differences in egg to fry survival due to oiling are detected. Average survival from unoiled areas will be used to estimate potential fry density in oiled areas. Observed and potential fry densities will then be expanded to estimate total observed and potential fry. The difference between the two estimates will be considered lost fry production.

Specific statistics to be estimated are:

1. Number of dead and viable eggs per square meter by salmon species, stream, and stream zone.
2. Number of dead and live fry per square meter by salmon species, stream, and stream zone.
3. Egg to fry survival by salmon species, stream, and stream zone.
4. Lost production by salmon species, stream, and stream zone.

SCHEDULES AND REPORTS

Date(s)	Activity
Oct 23 - Nov 3 1989	Egg digs in 8 study streams.
Nov 6 - Dec 17 1989	Data entry and preliminary analysis
Mar 12 - Mar 23 1990	Fry digs in same 8 study streams.
Mar 26 - May 3 1990	Data entry, final analysis, and report.

PROJECT BUDGET [1]

Line Item	Category	Budget
100	Personnel Services	$ 5,200
200	Travel	$ 600
300	Contractual	$ 5,000
400	Commodities	$ 800
500	Equipment	$ 4,100
700	Grants	$ 0
Total		$ 15,700

[1] Budget is for all activities performed from March 27, 1989 to February 28, 1990.

FUNDED PERSONNEL

Class	PCN	Name	PFT_mm	SFT_mm
FB II	11-1258		2.5 (funded under CF-381)	
FB I	new			1.5
FT III	11-1590			0.7
FT II	11-1369			0.7

II. KODIAK AND CHIGNIK MANAGEMENT AREA STUDIES

INTRODUCTION

During 1989, a number of commercial salmon fisheries in the Kodiak and Chignik Management Areas were closed as a direct result of the Exxon Valdez oil spill. A potential outcome of these closures is overescapement of pink and chum salmon in some streams. The goal of this investigation is to quantify effects of the 1989 escapements upon future brood year production. Pink and chum salmon constitute 87% (6,517,000 pinks, 745,000 chums) and 35% (705,097 pinks, 165,725 chums) of the Kodiak and Chignik salmon harvests, respectively. The average value of pink and chum salmon harvest during the last nine odd years (years of high pink salmon abundance) in Kodiak and Chignik has been 14.8 and 3.7 million dollars (Malloy 1989; Thompson and Fox 1989).

Within the Kodiak and Chignik Management Areas there are 346 streams that support populations of pink salmon and 90 streams that support populations of pink and/or chum salmon. Pink salmon pre-emergent sac fry sampling has been conducted in 43 Kodiak and 8 Chignik streams periodically over the last 20 years. Indices of pre-emergent sac fry abundance coupled with aerial survey escapement counts, provide the foundation for forecasting returns and projecting harvest potential of these species.

To evaluate potential damage to future brood year pink and chum salmon production, total escapements, pre-emergent sac fry abundance, available spawning habitat, and fecundity and egg retention variables can be used to evaluate trends in overwintering survival of egg to pre-emergent sac fry. A relationship using these objective measures for quantifying potential damage to future brood year production will be developed and assessed by the project.

OBJECTIVES

A. Determine abundance of pink and chum salmon eggs and pre-emergent fry. Specific objective includes:

   1. Derive a length-fecundity relationship for odd year Kodiak and Chignik pink salmon.

   2. Determine egg retention and fork length for female pink salmon in selected pre-emergent streams.

   3. Ascertain total available spawning habitat for selected pre-emergent index streams.

   4. Estimate potential egg deposition for pink and chum salmon in selected streams.

5. Estimate number of pre-emergent sac fry.

B. Determine overwinter mortality (egg to pre-emergent fry) of pink and chum salmon eggs.

C. Determine reductions, if any, in pink and chum salmon pre-emergent fry abundance due to oiling.

D. Identify potential alternative methods and strategies for restoration of lost use, populations, or habitat where injury is identified.

METHODS

Length-Fecundity Sampling

Personnel stationed at the four weirs (used for foot surveys and escapement counts in Fish Study Number 7) will also collect pink salmon length-fecundity data. One hundred female salmon will be randomly sampled (prior to upstream migration) from each wired system. An additional sample will be taken from a stream in the Chignik Area. The sample size (100 females per system) is a compromise between effort associated with sampling and number of samples necessary to determine the length-fecundity relationship. Each sampled fish will be killed, length will be measured to the nearest millimeter, and all eggs will be counted in the field. Sampling will be spaced over a two week period in an attempt to represent a wide spectrum of the particular run. All data will be recorded on form AKA89-2 (Appendix A.1).

Index Stream Survey and Spawner Distribution Mapping

Using enlargements (27% greater than 1:1) of 1:250,000 United States Geological Survey (U.S.G.S.) topographical maps encompassing index streams, experienced management personnel will demarcate upper and lower limits of pink salmon spawning distribution based upon aerial surveys from previous years. Information from aerial escapement surveys during 1989 (Fish Study Number 7) and spawner distribution derived from helicopter flights from this study will be integrated into maps to provide detailed spawner distribution information for all index streams. Spawner distribution for this study will be marked on maps during peak spawning activity (or noted otherwise) and aerial estimates of pink salmon numbers will be recorded by stream reach.

Egg Retention Sampling

From each index stream selected for surveying, 150 fish will be sampled from which to estimate egg retention. Sampling intensity will be proportional (weighted) to the level of spawning in a stream reach. Therefore, sampling will be concentrated in areas where major spawning densities have been noted by the aerial

escapement survey observers (Fish Study Number 7). Sampling will be conducted using a systematic stratified sampling design. In streams where major concentrations in specified areas are low (< 1,000 carcasses) every other female will be sampled, where densities are high (> 5,000), every fifth female will be sampled until the complete sample of 150 is obtained. Sampling procedures will consist of measuring each fish (mid-eye to fork-of-tail), making an incision in the fish, removing all eggs retained in the body cavity, and placing the eggs in a ziplock bag. Each sample will be labeled with the stream name, number, and length of the fish sampled. If an individual fish has fewer than 200 retained eggs, the eggs will be counted in the field. Data will be recorded on form AKA89-4 (Appendix A.2).

Spawning Habitat Survey Design

Using maps which delineate the upper and lower limits of spawner distribution for a given index stream, total stream length will be estimated in meters using a calibrated map wheel. Overall stream length (length of stream available to spawning fish) will be measured with no attempt made to stratify tributaries or braided channels, which will be included in total length measures. The entire stream will be divided into 300 meter sections beginning at the lower, and extending to the upper limits of historical spawner distribution. Each section will be assigned a random number beginning at 01, and sections to be sampled for spawning habitat surveys will be selected using a random number generator.

Numerous aquatic habitat inventory survey designs exist (Platts et. al. 1983; Frissell 1986; Murphy et. al. 1987; Hankin and Reeves 1988), however, for purposes of this study, a systematic cluster transect sampling design was deemed most appropriate based upon temporal constraints and unique requirements of the objectives. Five 300-meter long sections will be randomly selected to be sampled from each stream. Beginning at the downstream end of each of these sections, cluster transects will be run perpendicular to the stream bank at 25 meter intervals. Twelve transects per section and a maximum of 60 per stream will be measured.

According to the literature and also habitat suitability index models from Raleigh and Nelson (1985), substrate size and water velocity appear to be the most critical variables with regard to spawning success of pink salmon. Substrate embeddedness may also have an impact on spawning success (Platts et al. 1983), however its importance as a quantitative variable related to initial pink salmon life history stage survival is unknown. At each sample transect, stream width, spawning habitat, water velocity, depth, and substrate embeddedness will be recorded. Stream width will be measured along each transect from a minimum depth of six inches of water to the opposite stream bank or to the point where water is less than 6 inches in depth. In maintaining consistency of measures both within and between streams, protruding logs, rock outcroppings, etc. surrounded by water are included in measurements of width. Islands are not included; an island is any object

protruding above the water surface greater than 0.3 m in width. Ranges for substrate sizes, water velocity and depth were developed using an average of values in the literature (Divinin 1952; Chambers 1956; Andrew and Geen 1960; Krueger 1981; Neave 1966; Wilson et. al. 1981).

Available spawning habitat will be estimated based upon the previously mentioned variables and recorded as a percent. An area will be considered as suitable spawning habitat for pink salmon if substrate sizes are between 0.58 and 13.75 cm, stream depth is greater than 0.15 m, water velocities are between 0.28 and 0.9 m/second, and if stream embeddedness is such that excessive foot pressure must be exerted before gravel is displaced. Data will be recorded on form AKA89-3 (Appendix A.3). To standardize data collection, all personnel who will perform sampling will receive a 12 hour training session with experienced field personnel.

Pre-emergent Fry Sampling

Sampling of pre-emergent sac fry will be conducted on index streams and stream reaches in the Kodiak and Chignik Management Areas based upon results of Fish Study Number 7, finalized spawner distribution maps, escapement magnitudes and spawner densities. Selection of index streams and stream reaches to be sampled will be the responsibility of the Regional Biometrician. All streams and reaches in the Kodiak Area which are sampled for pre-emergent fry during normal sampling by the Commercial Fish Division will be treated separately from the present investigation. Circular digs (0.3 m^2) will be conducted along a transect line run perpendicular to the stream bank in selected areas. The number of digs per transect will vary depending on analysis of spawner density data. Digs will be conducted using a high pressure hose to extract eggs and fry from the substrate and a specially designed net will be used to catch eggs and fry as they are displaced. Numbers of live and dead fry and eggs by species will be collected and counted from each 0.3 m^2 dig. Data will be recorded on water resistant forms in the field and transferred to the regional data base upon completion of a specific stream.

DATA ANALYSIS

The analytical component of this investigation will integrate data and results from Fish Study Number 7 as well as historical escapement and pre-emergent sac fry indices, length-fecundity data, potential egg deposition and spawning habitat estimates. A cohesive series of analyses will address the following estimates:

1. Total available spawning habitat in index streams for Kodiak and Chignik.

2. Spawner density in No./m^2 for a spectrum of escapement magnitudes.

3. Potential egg deposition for pink and chum salmon for the index streams addressed.

4. Number of pre-emergent sac fry in index streams.

5. Overwinter mortality (egg to pre-emergent fry) in index streams.

SCHEDULES AND REPORTS

Dates	Activity
June - September	Field surveys, data collection
October - November	Historical data collection, editing, preliminary analysis and map preparation
December 1 - December 21	Data analysis, interim report submitted
December - February 1	Report Finalization
February 10	Pre-emergent fry sampling

PROJECT BUDGET

Line Item	Category	Budget[1]
100	Personnel	39,200
200	Travel	800
300	Contractual	44,100
400	Supplies	6,900
500	Equipment	6,900
700	Grants	-0-
Total		$95,700

[1]Budget includes activities performed from 15 July 1989 to February 1990.

FUNDED PERSONNEL

Class	PCN	Name	MM	OT	SEA	HAZ	COST
FB IV	1202	D. Schmidt	0.0	0	0	0	-0-
FB III	7016	B. Barrett	0.0	0	0	0	-0-
FB II	7017	C. Swanton	7.5	350	0	0	30,054.72

LITERATURE CITED

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CONFIDENTIAL

STATE/FEDERAL NATURAL RESOURCE DAMAGE ASSESSMENT
DETAILED STUDY PLAN

Project title:	Early marine salmon injury assessment for the Kenai Peninsula and Kodiak/Shelikof Strait
Study I D Number:	Fish/shellfish study no. 9
Lead Agency:	State of Alaska, ADF&G Fisheries Rehab, Enhancement and Development Division
Cooperating agencies:	
Principal Investigator:	Jim Raymond, Fishery Biologist
Assisting personnel:	Nick Dudiak, Lower Cook Inlet Lonnie White, Kodiak
Date submitted:	11 October 1989

DRAFT

	Signature	Date
Principle Investigator:	Jim Raymond	10/11/89
Supervisor:		
Consulting Biometrician:		
OSIAR Senior Biometrician:		
OSIAR Program Manager:	Richard Holmes	10/13/89
OSIAR Director:		

Note: The Fishery Management Team did not request field work to start on this study until 1990, and no field work has so far been done. This is an operational plan for development of a detailed study plan by February 1990 for the 1990 field season. It includes budgets for the periods 3/1/89-2/28/90 and 3/1/90-2/28/91.

INTRODUCTION

The early marine period is a critical one for salmon because it is during this period that the greatest mortalities are incurred. Reductions in food supply or access to rearing areas can reduce growth and prolong the period when the fry are vulnerable to predators.

Some estuarine and intertidal nursery areas important to salmon were heavily impacted by the oil spill. These include several bays in the Kenai Fiords area, in the lower Kenai Peninsula, and on Kodiak Island. It is anticipated that these impacts may have detrimentally affected the viability of salmon production in these areas and the resultant viability of the present fisheries and related economy.

In this study, oiled and unoiled paired sites will be selected in each area and investigated for effects of oil on both salmon fry and their food supply. This study will differ from Project 4 (injury to juvenile salmon within Prince William Sound) in that it will not include recoveries of coded-wire-tagged fish, since few tagged fish will be in the regions of study.

OBJECTIVES

A. Determine the impact of the oil spill on abundance, growth, diet and stomach fullness of fry.

B. Determine the impact of the oil spill on food supply, as measured by plankton and epibenthos abundance and composition.

C. Determine hydrocarbon levels in tissues of juvenile salmon in oiled and unoiled areas and document any oil-related mortalities and other adverse changes in viability.

D. Identify opportunities for restoration of lost habitat or fish where injury is identified.

METHODS

The following is only a brief outline of expected methods to be used. Detailed methods will be described in the study plan presently under development.

Each component will include pair-wise comparisons of oiled and unoiled sites similar to that conducted in Prince William Sound in 1989. Sites will be chosen that have similar gradient, substrate, exposure, and currents. All methods and techniques will be made compatible with those already in use in the early marine salmon study in Prince William Sound (Study No. 4).

1. Lower Cook Inlet. Nick Dudiak, Study Leader

Target species: pink and chum salmon juveniles

Three strategies will be used to assess effects of oil on juvenile salmon and their habitats: 1) A pairwise comparison of oiled and non-oiled habitats and the abundance, growth, feeding habits, behavior and food resources of juvenile salmon in these habitats; 2) Comparisons with historical data on juvenile salmon ecology in Lower Cook Inlet.

Three paired sites will be selected. Two pairs will be selected in the Outer District and one pair in the Southern District. Tutka Bay will be used as one "control", non-oiled area in the Southern District because historic juvenile salmon and food habit data are available.

Every other week from April - August, a crew of three in 16 - 18 foot skiffs will collect juvenile pink and chum salmon at these paired sites using beach and purse seines. Plankton and epibenthic sampling will also be conducted in conjunction with juvenile salmon sampling on an alternate week basis.

Fry samples taken at each site will be analyzed for CPUE, size, diet and stomach fullness. Periodically, fry samples will be collected and frozen for later archiving and possible hydrocarbon analysis.

Horizontal plankton and shallow epibenthic samples will be taken at all paired sites twice-monthly basis during the April - August sampling period. Oceanographic measurements, such as temperature, salinity, and physical sea conditions, will be recorded at each sampling site each period.

The level of hydrocarbon contamination of the tissues of juvenile salmon will be assessed by pairwise comparisons between oiled and non-oiled areas.

2. Kodiak Island. Lonnie White, Study Leader

Target species: pink and underyearling sockey salmon juveniles.

Study areas will include the Izhut and Raspberry Strait areas which have juvenile salmon and which were affected by oil. Both oiled and unoiled nursery areas are be found in these areas. Sampling

will be done between March 15 and August 15.

Sites will be visited each week. A 25-ft Boston Whaler will be used as the sampling platform and for transportation from Kodiak to the study sites.

Records will be kept of CPUE for three beach seine hauls at each site. A total of 30+ juvenile salmon will be preserved in formalin for growth information and 10 stomach samples will be taken for diet information. Two replicate plankton and epibenthic samples will be taken at each site.

Observations will be made of oil observed in the study areas. Fry will be pooled to obtain 10-gram tissue samples will be taken and archived for later hydrocarbon analyses.

3. Resurrection Bay-Kenai Fiords. Work in this area may be done under contract to another agency, possibly the USF&WS if the US Dept. of the Interior extends its activities beyond 28 February 1990. A recent baseline study by the USF&WS of salmon fry in Resurrection Bay will be used as a reference.

BUDGET SUMMARY

1. Present - 2/28/90

Area	100	200	300	400	500	Total	Details on page
Lower Cook Inlet	16.0	0.2	0.0	5.2	14.5	35.9	4
Kodiak/Shelikof	16.0	1.0	2.2	6.2	9.0	34.3	6
Kenai-Resurrection[a]						35.0	

[a]Inclusion not yet decided, estimated budget if approved.

2. 3/1/90- 2/28/91

Area	100	200	300	400	500	Total	Details on page
Lower Cook Inlet	74.2	0.5	51.8	0.8	4.5	131.8	5
Kodiak/Shelikof	75.0	1.5	32.6	13.2	5.6	127.9	7
Kenai-Resurrection[a]							

[a]Inclusion not yet decided

LCI BUDGET
Present - 28 February, 1990

LINE 100:

No.	Class	Name	Months	Salary/ Benefits	Total
7048M	FB I PFT	Boyle	4.0	4.0	$16.0

LINE 200:

1 Round trip to Anchorage @ $0.2	$0.2

LINE 300: -0-

LINE 400;

Miscellaneous sampling equipment: plankton nets, sampling containers, lab equipment	$3.2
Survival suits/float coats	2.0
Total	$5.2

LINE 500:

18 foot aluminum skiff, trailer, 50hp & 10hp motors	$11.0
D.O. and salinity meters with probes	3.0
Marine VHF radio	0.5
Total	$14.5
GRAND TOTAL	$35.9

BUDGET SUMMARY

Period: Present - 28 February, 1990

Task	100	200	300	400	500	Total
1. Site ID	16.0a/	-	-	-	-	$16.0
2. Literature survey	a/	-	-	-	-	-0-
3. Detailed project plans	a/	-	-	-	-	-0-
4. Order equipment & recruit personnel	a/	-	-	5.2	14.5	19.7
5. Interface w/ other studies	a/	-	-	-	-	-
6. Sample custody training	a/	0.2	-	-	-	0.2
			TOTAL			$35.9

a/ - FB I PFT position involved with all other tasks. Not broken down in this presentation.

LCI BUDGET ESTIMATE
1 March, 1990 - 28 February, 1991

LINE 100:

No.	Class	Name	Months	Salary	Total
7048M	FB I PFT	Vacant	12.0	4.0	$48.0
7049M	FT III S	Vacant	4.0	2.5	10.0
7034M	FT II S	Vacant	4.0	2.2	8.8
7052M	FT II S	Vacant	4.0	2.2	8.8
		Total:	24.0		$74.2

LINE 200:

1 round trip Anchorage/Cordova @ 0.5 — 0.5

LINE 300:

Charter support for sampling:

Vessel = $2.5/day x 4 days/months x 4 months	=	$40.0
Air = Otter @ $0.45/hr x 2hr/trip x 2 trips	=	1.8
Limnology Section lab analysis costs	=	10.0
Total		$51.8

LINE 400:

Miscellaneous sampling equipment: plankton nets, sampling containers, lab equipment, food — 0.8

LINE 500:

Survival gear, float coats, tools for boat, tide meter — 4.5

TOTAL $131.8

KODIAK BUDGET
Present - 28 February, 1990

LINE 100:

No.	Class	Name	Months	Salary/ Benefits	Total
7048M	FB I PFT	Honnold	4.0	4.0	$16.0

LINE 200:

3 Round trip to Anchorage @ $0.24 + $80 PD		1.0

LINE 300: -0-

Air charter site selection 10 hr @ @$200	2.0	
Phone and xerox	0.2	
Total		2.2

LINE 400:

Survival suits/float coats	1.35	
Safety equipment	0.5	
Beach seines 2 @ 850	1.7	
Plankton supplies	0.5	
chain of custody sample bottles	0.2	
office supplies	0.5	
photo supplies	0.5	
lab supplies	0.2	
purse seine	1.2	
dip nets	0.2	
chest waders, neoprene 2 @ $150	0.3	
Total		$6.15

LINE 500:

Yamaha outboards, 130 HP, 2 @ $3800	7.6	
D.O. and salinity meters with probes	0.9	
Marine VHF radio	0.5	
Total		$9.0
GRAND TOTAL		$34.4

KODIAK BUDGET SUMMARY
Present - 28 February, 1990

Task	100	200	300	400	500	Total
1. Site ID	4.0	0.0	2.0	0.0	0.0	6.0
2. Literature survey	2.0	0.0	0.0	0.0	0.0	2.0
3. Detailed project plans	4.0	0.4	0.2	0.0	0.0	4.6
4. Order equipment & recruit personnel	2.0	0.0	0.0	6.2	9.0	17.2
5. Interface w/ other studies	2.0	0.3	0.0	0.0	0.0	2.3
6. Sample custody training	2.0	0.3	0.0	0.0	0.0	2.3
	16.0	1.0	2.2	6.2	9.0	34.4

KODIAK BUDGET ESTIMATE
1 March, 1990 - 28 February, 1991

LINE 100:

No.	Class	Name	Months	Salary	Total
7048M	FB I PFT	Honold	12.0	4.0	$48.0
7049M	FT III S	Vacant	4.0	3.5	14.0
7034M	FT II S	Vacant	4.0	3.0	12.0
		Total:	20.0		$75.0

LINE 200:

3 round trip Kodiak/Anchorage @ 0.5		1.5

LINE 300:

Aircraft/boat charter	15.0	
Limnology Section lab analysis costs	15.0	
Phone 12 @ $50	0.6	
motor repairs	2.0	
Total		$32.6

LINE 400:

Gasoline 2000 gal @ $1.20 and oil $200	2.6	
Groceries 600 man-days @ $11	6.6	
Replacements for items shown in previous year	4.0	
Total		13.2

LINE 500:

Avon raft, 14-ft	3.6	
Outboard, Yamaha, 40 HP	2.0	
Total		5.6
TOTAL		$127.9

STATE/FEDERAL RESOURCE DAMAGE ASSESSMENT
DETAILED STUDY PLAN

Project Title:	INJURY TO DOLLY VARDEN CHAR AND SOCKEYE SALMON IN THE LOWER KENAI PENINSULA
Study ID Number:	Fish/Shellfish Study Number 10
Lead Agency:	State of Alaska, ADF&G Sport Fish Division
Cooperating Agencies:	Federal: USFS State: DNR
Principal Investigators:	Kelly Hepler, Fishery Biologist Nicholas Dudiak, Fishery Biologist
Assisting Personnel:	Andy Hoffmann, Fishery Biologist 6 Fishery Technicians
Date Submitted:	September 25, 1989

APPROVED

Titles	Signature	Date
Principal Investigator:	Kelly Hepler	10/9/89
Supervisor:	Doug Mecum	10/9/89
Principal Investigator:		
Supervisor:		
OSIAR Senior Biometrician:		
OSIAR Program Manager:	Ronald Holmes	10/13/89
OSIAR Director:		

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INTRODUCTION

The goal of this project is to determine whether the Exxon-Valdez oil spill will have a measurable impact on anadromous stocks of Dolly Varden char, *Salvelinus malma*, (hereafter referred to as char); and, sockeye salmon, *Oncorhynchus nerka*, (hereafter referred to as sockeye) of the lower Kenai Peninsula (LKP).

Char are estuarine anadromous species that inhabit much of LKP. Unlike anadromous Pacific salmon, *Oncorhynchus sp.*, char utilize near-shore and estuarine areas for feeding and their marine migrations are not as extensive as those of Pacific salmon (Armstrong and Morrow 1980). Stocks of this species inhabit areas that have been severely affected by direct contact with oil including Delight and English Bay Lakes. Since this species commonly live to age 8 (Morrow 1980), the potential exists for long-term effects from exposure to oil. Study of this species is crucial in that it represents the only finfish species (with the exception of cutthroat trout, *Oncorhynchus clarki*) in the fishery assessment proposal package that inhabits the most oil-affected areas (the near-shore waters of the LKP) throughout most of it's life.

The lower Kenai Peninsula stocks of sockeye are important to the sport, commercial and subsistence fisheries in lower Cook Inlet. Anadromous sockeye utilize nearshore marine regions during the early stages of their marine residence, feeding on zooplankton, insects, and small fishes (Morrow 1980). Stocks of sockeye from Delight and English Bay Lakes inhabit marine areas that have been severely affected by direct contact with oil, however many young fish that emigrate from Leisure Lake and Big Lake will not have had contact with oiled nearshore marine areas. The potential impact on survival of sockeye while inhabiting the oil-affected areas (near-shore waters of the lower Kenai Peninsula) requires assessment of smolt production and adult returns (harvest and escapement) to these systems.

Life History

Dolly Varden Char:

The experimental design for the char portion of this program is based upon the life history model developed by Armstrong (1970, 1974, 1984; Armstrong and Morrow 1980) to explain the migratory behavior of anadromous char. This model identifies two patterns of life history: fish that were spawned in lake systems, and fish that were spawned in non-lake systems. For both groups, juvenile char remain in freshwater residence in their natal stream for up to four years. During their last spring of freshwater residence, they smolt to sea. During late summer or early fall, fish that were spawned in lake systems then return to their natal stream to overwinter in the freshwater lake. During the spring, they emigrate into marine waters and annually return to their natal lake system during late summer or early fall to spawn and overwinter. Fish that were spawned in non-lake systems exhibit a more complex migration. Upon smoltification, juvenile char search for a lake

system to overwinter. These fish then behave in the same manner as do fish that originate in a lake system except that the return to their natal stream to spawn, and then return to their lake system to overwinter.

Sockeye Salmon:

Sockeye salmon are anadromous fish that spawn primarily, but not exclusively, in streams associated with lakes. The fry spend their first or second year rearing in lake habitat and smolt the following spring. Young sockeye stay close to shore and move out to the open ocean as they grow larger. After two years in the marine environment they return to their natal streams for spawning (Morrow 1980).

Experimental Design

Dolly Varden Char:

It is hypothesized that two detrimental impacts on this species could result from the presence of large amounts of crude oil in marine waters including: (1) reduced survival; and (2) reduced growth. To test whether there will be measurable impact on these stocks, one or two lake systems having stocks of char from each of two treatments were selected for study. A high-impact treatment is defined as a stock emigrating from a lake system which flows into a marine environment which has been directly impacted by oil, while a low-impact treatment is defined as stocks exiting lake systems into areas which have not been directly impacted by oil.

The principal objective of the project is to measure annual abundance, survival, recruitment, and growth of the stocks of char in each of the study streams over three calendar years. Our primary assumption is that there is a difference in exposure to oil for fish stocks from each of the two treatments. Evidence from the literature indicates that marine migrations can range up to 116 kilometers (Armstrong 1974). In addition, we will also test whether fish that enter systems tributary to non-oiled marine waters migrate into oiled systems to overwinter; thus providing a measure of the extent of fish exposed to oil. Although, marine migrations from low-impact waters may extend into oiled marine waters, the stocks selected within each treatment still represent different treatments in that the marine waters first encountered upon entry from freshwater will be very different in their oil content. Thus, it is assumed that any significant changes in stock abundance, composition, dynamics, or tissue composition from the initial emigration of stocks in the oiled treatment as compared to stocks from the non-oiled treatment is due to contact with the oiled marine waters.

Armstrong's model of migratory behavior provides the basic framework for this study. Each of the study lake systems represents a stock of fish that annually homes to that specific overwintering stream. Although sampling will not be conducted until spring 1990 (which is one year after the original oil contamination), fish older than age 5 would have

been present in their respective lake system during the 1989 emigration. Also, we expect oil to persist in these marine waters and testing of these fish in subsequent years after contamination remains a valid approach. In addition, a non-oiled non-lake system that supports a large number of char will be sampled to measure whether these fish migrate into lake systems tributary to oiled marine waters to overwinter. Since this system does not contain a lake, these fish will migrate to a lake system to overwinter (Armstrong 1984).

A measurable detrimental impact on these anadromous stocks of char may result in a loss to the sport fishery. The status of the sport fishery will be investigated through: (1) an ongoing mail questionnaire (Mills 1988); (2) an ongoing creel survey and stock status program for the Anchor River, Appendix B (Larson 1988); and (3) a creel survey of selected LKP marine ports (OSIAR study FS# 6).

Sockeye Salmon:

It is hypothesized that the presence of large amounts of crude oil in marine waters could reduce the survival of marine sockeye smolt. To test this hypothesis four study sites were chosen. Two of these, Delight and English Bay lakes, were directly affected by the oil spill and two Big Lake and Leisure Lake were not.

The strategy is to measure the marine survival rate for each age class of sockeye salmon and compare the survival rates of the oil impacted areas to those that were not affected by the oil.

OBJECTIVES

During 1990, the specific objectives of this project are:

Dolly Varden Char:

1. to test the hypothesis that there is no difference in annual survival rates of char between oiled and non-oiled lake systems (the test will be done given a level of significance of $\alpha = 0.05$);

2. to test the hypothesis that there is no difference in annual growth rates of char between oiled and non-oiled lake systems (the test will be done given a level of significance of $\alpha = 0.05$); and,

3. to test the hypothesis that char from non-oiled, non-lake systems migrate into oiled systems (the test will be done given a level of significance of $\alpha = 0.05$).

4. to identify potential alternative methods and strategies for char restoration of lost use, populations, or habitat where

injury is identified. (To be accomplished at project completion).

Sockeye Salmon:

5. to test the hypothesis that there is no difference in annual survival rates of sockeye between oiled and non-oiled lake systems (the test will be done given a level of significance of $\alpha = 0.05$);

6. to identify potential alternative methods and strategies for sockeye restoration of lost use, populations, or habitat where injury is identified. (To be accomplished at project completion).

To accomplish these objectives the following tasks will be performed.

Dolly Varden Char:

1. Count the char emigrating in spring through weirs placed on three lake systems, two in oiled areas and one in a non-oiled area.

2. Estimate annual survival rates for each of the overwintering study stocks using abundance data collected at the weirs.

3. Count the char immigrating into a non-lake system in late summer; but emigrating from this non-lake system during the fall.

4. Estimate mean length of emigrating trout and char from each study stream (the estimates will be ± 10 mm of their true values 90% of the time).

Sockeye Salmon:

5. Estimate the number of smolts emigrating in the spring through weirs placed on four lake systems, 1990

6. Count adults immigrating through the weirs in the summer (escapement) on four lake systems, 1991-1992.

7. Estimate the proportions of harvested adults by age such that each estimate will be within ± 5 percentage points of the true proportion 95% of the time, 1991-1992.

8. Estimate the proportions of escapement by age to the four lake systems such that each estimate will be within ± 5 percentage points of the true proportion 95% of the time, 1991-1992.

9. Tally the harvest of adults in terminal fisheries on stocks from three lake systems, 1991-1992.

10. Estimate the harvest of adults in the mixed stock fisheries of Cook Inlet on one stock such that the estimate is within ± 5 of the true harvest 95% of the time, 1991-1992.

11. Estimate the mean length of adults (1991 and 1992) and smolts (1990) such that each estimate is within 5 mm 95% of the time.

METHODS

Study Design

Dolly Varden Char:

The study includes four systems; English Bay and Delight Lake, which are in areas exposed to oil, and Packers Lake and Anchor River, which are in non-oiled areas. The first three are systems with lakes on them, while Anchor River does not contain a lake in its drainage.

Based on Armstrong's migratory model; all spring emigrants from the three lake systems will return, overwinter, and emigrate the following spring which allows for estimation of annual survival and growth for the population. These systems will be weired in the spring and all emigrating char will be counted, sampled for length, tagged and examined for tags from previous years. The hypothesis of equal annual survival and growth will be tested on these three systems, with two representing the treatment (oiled) and one being the control.

Char emigrating from the non-lake system (Anchor River) will search for and overwinter in a lake system; only returning to their natal stream for spawning (Armstrong 1984). Since the char from the Anchor River will emigrate to other systems to overwinter this system will be weired during both the summer and fall. All char migrating upstream and downstream will be counted and examined for tags from previous tagging; length samples will be taken; and all emigrating, spawning char will be tagged. The details of this program are presented in an ADF&G Sport Fish Division Federal Aid in Fish Restoration Operational Plan, Appendix B (Larson 1989).

Survival Rates. Survival will be estimated for smolts, immature and mature char. Initially, all char up to 200 mm will be considered smolt. All char between 200 mm and 250 mm will be considered to have been immature the year before. Blackett (1968) found average length of mature char to be in the range 250 mm and larger and the smallest spawning char sampled in Kodiak in 1988 were at 250 mm. Char over 250 mm in length will be classified as mature. At the conclusion of the year's sampling, we will analyze length frequency data to identify size classes of smolt.

Char over 200 mm will be tagged with numbered Floy FD-68 anchor tags. Char under 200 mm and down to 110 mm will be tagged with numbered Floy

Fabric tags. If all fish can be censused and examined for tags in all years, survival will be known for each system. Annual survival will be estimated for three groups, smolts, immature and mature char. The mortality rate of spawning char is known to be high, particularly for males (Armstrong 1974), so the rate of survival estimated for smolt and immature char will be used to test the hypothesis of equal survival between oiled and non-oiled systems.

The hypothesis of equal survival will be tested using a chi-square statistic. The goals set in Table 1 for number of fish to mark are calculated under the assumption that all char are censused and examined for tags. However, if unknown numbers fish can be expected to be lost past the weir, then survival cannot be estimated directly from the numbers released and returned, instead survival will be estimated with mark-recapture methods. Estimates of survival (Seber 1982) from a mark-recapture experiment with their 95% confidence intervals at three levels of abundance were examined to estimate the sample goals required to detect significant differences in survival (Appendix A). The average emigration of char from Packers Lake for the past 6 years has been at 30,000 fish, ranging from 6,000-40,000. Information from Armstrong (1974) indicates that survival of smolt to spawning is 10-15%. An expected abundance of 30,000 fish and expected survival of 10%, is therefore used to set a minimum sample goal of 12,000 fish tagged and the percent of the emigration that should be examined for tags (Table 2). However, this is a fall-back position, a minimum which must be achieved if all fish cannot be handled due to external, uncontrollable factors.

At the tagging level of 12,000 fish, and if all fish are examined during the second and third springs, the test for independence will detect a difference in survival as small as 1-2% with 95% confidence (Table 1).

No information is available on what numbers can be expected to emigrate from English Bay or Delight Lake. However, historical data from Packers Lake and data from char projects in Kodiak and Prince William Sound indicate that an average migratory timing function could be estimated for emigrating char. This function will be used to expand the numbers emigrating at each weir during the early stages of the migration, to estimate the total emigration. If this rough expansion indicates that the final numbers emigrating will be much higher or lower than 30,000 fish, then the total to be tagged and the minimum percent to examine will be adjusted using Table 2.

A subsistence fishery occurs in the English Bay system above the weir site and estimation of this source of fishing mortality will be necessary. We will work with participants in this fishery in an attempt to recover all tags harvested in the fishery. Assuming that the marked-to-unmarked ratio in the fishery is equal to the subsequent emigration, total harvest can be estimated by expanding total tags harvested by this ratio. Survival estimates for English Bay will then be adjusted for the fishery mortality.

Estimates of survival will be adjusted, if necessary, for differential fishing mortality from the sport fishery. Fishing mortality for the various tag lots will be estimated using the methods of Clark and Bernard (1987). Examination of sport-caught fish for tags will be accomplished through the port sampling program conducted as part of the Prince William Sound and Gulf of Alaska Sport Fishery Harvest and Effort Project (OSIAR study FS # 6). Estimates of sport harvest will be obtained from an ongoing postal survey (Mills 1988).

Growth. The hypotheses of equal growth between oiled and non-oiled sites will be tested by analysis of individual growth rates. Incremental growth for individuals will be computed from recaptured fish. The experiment will be conducted as an Analysis of Variance between the three lake systems. Years, areas and possibly initial length, will be factors in the design.

Migration. In order to test the hypothesis that Anchor River char migrate to the oiled systems to overwinter, all emigrating spawners will be tagged in the fall. These fish will subsequently be recovered at the weir on the lake systems. In 1988, 15,000 char larger than 200 mm migrated upstream in the Anchor River and by October 13th only 2,000 had migrated downstream (Larson in press). Assuming that the majority of those remaining upstream were spawners, and assuming spawning mortality to be 50%, then approximately 6,500 spent char would have left the system in 1988. During 1989, a floating weir will be installed in the Anchor River to allow for weir operation through mid-November. This should be sufficient time to enumerate and tag all fish that will emigrate from the system to an overwintering site (Armstrong and Morrow 1980).

Sockeye Salmon:

Survival rates: Survival rates of smolt emigrating through oiled estuaries will be compared to survival rates of smolt emigrating through unoiled areas. Stocks from English Bay and Delight Lake were chosen to be representative of "oiled" stocks while sockeye from Leisure Lake and Big Lake (Fish Creek) were chosen to be representative of "unoiled" stocks. Smolts leaving these watersheds in 1990 will return in 1992 and 1993 as adults. The first three stocks are located on the southern tip of the Kenai Peninsula (Figure 1); they were chosen because they are the largest stocks of sockeye salmon in their immediate area with terminal fisheries. Big Lake is north of Cook Inlet with a stock exploited in a mixed-stock fishery. However, the unique scale patterns of these fish can (and are) used to estimate the number of these fish caught in the mixed-stock fishery (Cross and Goshert 1988).

Survival rates of sockeye salmon will be estimated as the ratio of returning adults to emigrating smolt. New programs (weirs) to estimate the number of smolt will be established in 1990 and continued through 1991 at English Bay and Delight Lake (these programs will also be used

to count char). Numbers of smolt emigrating from Big Lake and Leisure Lake will be estimated with programs currently in place with modifications to assure consistency with described for English and Delight lakes (Bectol and Dudiak, 1988; Kyle in press) (weirs). New programs will be established in 1992 and continued through 1993 to count escapement of adults into English Bay and Delight Lake (weirs). Escapement into Big Lake will be counted through a program currently in place (a weir) with modifications to assure consistency with described for English and Delight lakes (unpublished report, Chlupach). Since there is no escapement at Leisure Lake (the outlet is blocked to upstream migration), the harvest in the commercial, sport, and personal-use fisheries in the area will be the return (Yuen 1988). Fisheries for sockeye returning to English Bay and Delight Lake are terminal fisheries, making the return of sockeye the harvest plus the escapement. Although sockeye returning to Big Lake are caught in mixed stock fisheries in Cook Inlet, their catch in these fisheries can be estimated with good precision through an ongoing project (Cross and Goshert 1988).

Since the smolt emigrating in 1990 will return in 1992 and 1993, age composition of emigrating smolts and returning adults will be estimated through the counting of annuli on scales. According to sampling procedures to estimate multinomial proportions (Thompson 1987) and after adjustment for typical rates of scale regeneration, 424 smolts and 561 adult sockeye should be sampled during each sampling event to attain objective criteria. Since there are typically temporal changes in age composition of a sockeye stock as it migrates past a fixed point, there will be one sampling event during each week of each migration. Emigrating smolts and escapement of adults will both be sampled at weirs. Harvests of adults will be sampled in each terminal fishery (Leisure Lake, English Bay, and Delight Lakes) through ongoing catch sampling programs in Homer (Yuen 1988) and of the mixed stock fisheries farther north (Big Lake) (Cross and Goshert 1988).

The mean length of emigrating smolts and returning adults will be estimated for use as covariates with influence on survival rates and as a means to estimate harvest in numbers of fish. All smolts and adults sampled for later age determination will be measured from mideye to the fork of their tail. Since one sample of 424 fish is sufficient to meet objective criteria when SD = 53 mm (according to procedures in Cochran 1977), objective criteria for estimating mean length should be met without supplementing the sample.

Numbers of smolt will be estimated with a weir and fyke-net system with a sampling device in the codend. All smolts will be caught and counted during periods of low rates of migration; the sampler will be used only when smolts are too numerous to count without causing mortalities. During sampling, all emigrating smolt will be captured and counted; smolt emigrating at other times will pass unhindered through the weir. To better conform with the diurnal frequency of smolt migration, each day of sampling will be divided into "light" (0300-2100 hrs) and "dark" (2101-0259 hrs) strata. The light stratum contains thirty 36-min sampling periods, three of which will be randomly selected for sampling.

The dark stratum contains eighteen 10-min periods, twelve of which will be systematically chosen at intervals of 30 min with the first chosen randomly from periods between 2100-2130 hrs. Smolt programs should occur from approximately 1 May to 1 July.

Data Collection and Reduction

Dolly Varden Char:

The data collection will be divided into two seasonal periods: spring and summer-fall. During the spring sampling, weirs will be used to count the emigration of char from study streams and to collect biological samples. Weirs will be installed on Delight and English Bay Lakes in conjunction with sockeye salmon smolt projects which are addressed under a separate operational plan. Since the weirs will be capable of capturing sockeye salmon smolt, they will also be able to intercept all emigrating char. The weir program on Packer's Lake will be accomplished in conjunction with Cook Inlet Aquaculture Association (CIAA). CIAA already operates a sockeye salmon smolt weir during the spring and we will augment their crew to accomplish this work. The weirs will be operated from early May to mid-July.

During the summer-fall, sampling will occur on the Anchor River. This system will be weired during the period 1 July to 15 November. This portion of the program is an ongoing project of the Alaska Department of Fish and Game and details of the project are addressed in a separate operational plan, presented in Appendix B (Larson 1989).

All species moving downstream or upstream will be identified, counted, and the tag number of any tagged fish will be recorded. Date, sex (if identifiable from external maturation characteristics), and length (tip-of-snout to fork-of-tail to the nearest mm) will be recorded on mark-sense forms.

All data will be recorded on standard Division of Sport Fish mark-sense forms. All completed forms will be visually scanned for errors and corrected as necessary. Corrected forms will be sent to RTS in Anchorage for processing by optical scanning. Resultant data files and summary printouts will be checked for errors and corrected as necessary. Data associated with uncorrectable errors will be deleted from the data file. Corrected data files will be returned to RTS for archiving.

Sockeye Salmon:

The data collection for the sockeye tasks will be divided into two seasonal periods, spring and summer-fall; as well as being dispersed among various division's ongoing projects. New weir operations will be established at Delight and English Bay Lakes for monitoring immigration and emigration and collection of AWL data from sockeye smolts and adults as well as char. Smolt emigration and adult immigration data from Big

Lake will be collected by FRED Division personnel as part of their on going studies with modifications to assure consistency with that described for English and Delight lakes (unpublished reports, Chlupach). Smolt emigration and adult immigration data from Leisure Lake will be collected using fyke nets or wier by FRED personnel as part of their on going studies (Bechtol and Dudiak, 1989), supplimented by OSAIR as needed. Harvest data for all four sites will be collected as part of the Division of Sport Fish's Statewide Harvest study for sport and personal use fisheries (Mills 1988) and the Commercial Fisheries Division's catch sampling program (Yuen 1988)

At weir locations, all species moving downstream or upstream will be identified, counted, and the tag number of any tagged fish will be recorded. Date, sex (if identifiable from external maturation characteristics), and length (tip-of-snout to fork-of-tail to the nearest mm) will be recorded on mark-sense forms. Scale samples will be collected for age determination.

All data from English Bay and Delight lakes will be recorded on standard Division of Sport Fish mark-sense forms. All completed forms will be visually scanned for errors and corrected as necessary. Corrected forms will be sent to RTS in Anchorage for processing by optical scanning. Resultant data files and summary printouts will be checked for errors and corrected as necessary. Data associated with uncorrectable errors will be deleted from the data files; and corrected data files will be returned to RTS for archiving. Data collected for Big Lake and Leisure Lakes in conjunction with other division's projects will be handled in one of two ways. If the data is collected for their objectives, the data will be handled through the respective division's standard data handling procedures then forwarded to us. If the data is collected exclusively for this project it will be processed as decribed above for Division of Sport Fish.

Data Analysis

Dolly Varden Char:

Estimates of annual survival will be computed through analysis of tag returns. If all emigrating fish can be examined for marks, the estimate of annual survival (S) can be simply computed as:

$$S = m_2 / R_1$$

where:
m_2 = number of fish recovered in year y+1
R_1 = number of fish tagged in year y.

The Jolly-Seber three-sample method (Seber 1982) will be used in the event that each emigrating fish cannot be examined at the weirs. Buckland's program RECAP (1980) will be used to generate the estimates and variances.

The assumptions of the Jolly-Seber model are:

1. every fish, tagged and untagged, has equal probability of being caught;

2. every tagged fish has equal probability of survival to the next sampling event;

3. every fish sampled has equal probability of being returned to the population;

4. there is no tag loss; and,

5. all samples are instantaneous.

The sampling event for the purposes of the mark-recapture experiment is the emigration of char past the weirs, as all emigrating fish must cross the weir and so are assumed to be equally vulnerable to being sampled. The assumption of equal survival of tagged fish will be tested for the different tag groups and for different length classes using chi-square statistics. Tag loss will be estimated for fish tagged in 1989, as all tagged fish will also be adipose fin-clipped. The last assumption is technically violated as the sampling event lasts through the entire migration. However, as each fish will only pass the weir once, and can only be sampled once, the emigration can be treated as an instantaneous sample of the whole population.

The hypothesis of equal survival will be tested using a chi-square test for independence. However the contingency table cannot be built if all char are not sampled at the weir during emigration during the second and third year of the experiment. In this case the 95% confidence intervals of the survival estimated from the multi-year mark-recapture experiments will be compared to test for significant differences. In order to examine the effect of initial length on subsequent survival; the tests and estimates will be stratified by tagging length, and if possible, a logistic regression will be used to estimate this effect.

Annual individual growth will be calculated from the tag data as the difference between length at time of release and length at time of recovery. An Analysis of Variance will be used to test for significant differences in growth between fish from oiled and non-oiled areas. Variation due to differences in years and initial length can be controlled for through the use of a block and covariate in the linear model if necessary.

The assumptions of Analysis of Variance are:

1. random sample,

2. normal distribution, and,

3. homogeneity of variance.

The assumption of normality will be tested using Kolomogorov's D statistic. In all likelihood the data will not be normally distributed and a logarithmic or a rank transformation will be necessary.

The homogeneity of variance assumption will be tested with a Barlett's test. Again, if the assumption is not valid a transformation will be used.

The rate of migration of char from the Anchor River into the three lake systems will be estimated as a binomial proportion.

Sockeye Salmon:

Estimated survival rates of sockeye salmon will be the ratio of the estimated abundance of returning adults to the abundance of emigrating smolt. Escapement and harvest of age 1.2 salmon in 1992 and age 1.3 salmon in 1993 will be summed to estimate the return from smolt emigrating in 1990. Since escapement and harvest of sockeye salmon by age will be estimated with independent programs, summing the variances will provide the variance of the sum. Resampling methods (Effron 1982) will be used to approximate the empirical distributions of harvest and return by age, of escapement by age, and of abundance of emigrating smolt. Monte Carlo simulation (Rubenstein 1981) will then be used on these empirical distributions of the numerator and denominator that comprise the survival rates to generate an empirical distribution for the survival rates themselves. Finally, these empirical distributions of survival rates will be compared to determine if there are significant differences among oiled and unoiled stocks.

The number by age of sockeye salmon harvested in terminal, commercial fisheries (English Bay, Leisure Lake, and Delight Lake) will be estimated as the product of the proportion of the sample for each age class and the estimated harvest in numbers:

$$\hat{H}_a = \hat{p}_a \hat{H}$$

$$V[\hat{H}_a] = \hat{V}[H]\hat{p}_a^2 + \hat{V}[\hat{p}_a]\hat{H}^2 - \hat{V}[\hat{p}_a]\hat{V}[H]$$

$$\hat{H} = H_w/\bar{w}$$

where:

H = harvest in numbers;

H_w = harvest in weight;

p_a = proportion of our sample of age a;

H_a = proportion of the harvest of age a; and

$\bar{w}$ = mean weight of a harvested fish.

Harvest as biomass will be obtained from regular fish tickets. Mean weight of the average sockeye salmon will be estimated with a standard length-weight regression for sockeye salmon. Enough length samples will be taken to lower the variance to near zero levels.

The number by age of sockeye salmon harvested in sport and personal-use fisheries (Leisure Lake) will be estimated as the product of the proportion of the sample for each age class and the estimated harvest in numbers as described above. Estimates of harvest in numbers will be obtained from an ongoing program of mail-in surveys of sport fishermen (Mills 1988). Since harvest is estimated as well as proportion by age, the estimated variance is that for products (Goodman 1960):

$$\hat{V}[\hat{H}_a] = \hat{V}[\hat{H}]\hat{p}_a^2 + \hat{V}[\hat{p}_a]\hat{H}^2 - \hat{V}[\hat{p}_a]\hat{V}[\hat{H}]$$

The number by age of sockeye salmon harvested in the mixed-stock fisheries in Cook Inlet (Big Lake) will be estimated as the product of the proportion of the sample for each age class and the estimated harvest from Big Lake:

$$\hat{H}_a = \hat{p}_a \hat{q} \hat{H}$$

$$\hat{V}[\hat{H}_a] = \hat{H}^2(\hat{V}[\hat{q}]\hat{p}_a^2 + \hat{V}[\hat{p}_a]\hat{q}^2 - \hat{V}[\hat{p}_a]\hat{V}[\hat{q}])$$

where q = the fraction of the harvest from Big Lake. Estimates of q will be obtained for an ongoing program of stock separation based on scale pattern analysis (Cross and Goshert 1988).

Numbers by age in the escapements will be calculated similarly to numbers by age in commercial fisheries in that adults will be counted at all weirs.

Since age compositions of the migrating populations of both smolt and adults change with time, estimates above will be stratified through time as suggested in Cochran (1977). Since no escapement can occur at Leisure Lake (the outlet is blocked to upstream passage), no stratification should be necessary as all fish mill near the stream.

SCHEDULES[1]

A schedule of tasks to be completed during 1990 is as follows:

Task	Date
Weir Operation (Anchor River Only)	7/01-11/15/90
Summer and Fall Sampling (All Areas)	5/01-9/15/90
Data Analysis and Report Preparation	9/15/90-2/15/91

[1]Note: Adult weirs will be in place during fall 1992 and 1993

REPORTS

Results of these study efforts will be reported to the Division of Oil Spill Impact Assessment and Restoration. Upon completion of litigative concerns, these data will be published as either an Alaska Department of Fish and Game, Sport Fish Division, Fishery Data Series report or in the fisheries literature.

BUDGET SUMMARY

A line item breakdown of the time period April 1, 1989 to February 28, 1990.

Line Item	Category	FY90
100	Personnel	84.0
200	Travel	0.7
300	Services	16.5
400	Commodities	45.0
500	Equipment	6.4
700	Grant	0
	Total	152.6

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Table 1. Number of Dolly Varden char smolts that need to be marked to test the hypothesis of equal survival, assuming spring emigration is 30,000 smolt. The assumption is made that all emigrants can be examined for tags in order to classify mortality versus survival.

From a Survival of	Given an alpha level of	To detect a difference of:				
		5%	10%	20%	25%	50%
15%	0.01	30,000	10,500	3,000	1,200	300
	0.05	19,500	6,000	1,500	900	300
	0.10	10,500	3,000	750	750	300
10%	0.01		10,500	2,100	1,500	300
	0.05	19,500	6,000	1,500	900	300
	0.10	10,500	3,600	900	600	300
5%	0.01		18,000	4,500	3,000	600
	0.05		10,500	3,000	1,500	300
	0.10	30,000	7,500	1,800	1,200	300

Table 2. Number of Dolly Varden char that need to be marked and the minimum needed to be examined for marks in order to estimate survival in a 3-sample Jolly-Seber experiment (refer to Appendix A).

Mean Abundance	Survival	Minimum Number of New Tags to Deploy	Minimum Number of Fish to Examine	95% C.I. (percent)	Relative Precision
10,000	5%	8,500	75%	0.036-0.064	57
	10%	8,500	75%	0.071-0.129	29
	20%	8,500	75%	0.171-0.229	15
	50%	8,500	75%	0.471-0.529	6
30,000	5%	12,000	50%	0.030-0.066	33
	10%	12,000	50%	0.080-0.102	17
	20%	12,000	50%	0.170-0.221	10
	50%	12,000	50%	0.379-0.521	4
50,000	5%	15,000	50%	0.032-0.068	36
	10%	15,000	50%	0.082-0.118	18
	20%	15,000	50%	0.181-0.219	9
	50%	15,000	50%	0.467-0.533	7

Appendices

Appendix A. Calculation of sample sizes for estimating survival.

Appendix A. Calculation of sample sizes for estimating survival.

A major objective of the study is to estimate survival and test the hypothesis that there is no significant difference between oiled and non-oiled areas. If all fish can be examined at all weirs in the second year of the experiment, then this hypothesis will be tested using a contingency table and a chi-square statistic. Sample sizes for marks needed to achieve a test of size 0.05 were estimated by running the analysis based on different survivals and changes in survival (Table 1). In order to be able to perform this type of test, the goal of the projects will be to examine all fish emigrating, while the number tagged will be determined as described below.

If fish are able to pass the weir unaccounted for, e.g. during a flood, then the above method cannot be used, and we will need to estimate survival using a multi-year mark-recapture method. A three-sample Jolly-Seber experiment will provide an estimate of survival from the first to the second year, θ_1 (Seber 1982). The equation for estimating this survival is as follows:

$$\theta_1 = \left\{ \frac{R_2 \, z_2}{r_2} + m_2 \right\} / R_1$$

and its variance is:

$$Var(\theta_1) = \theta_1^2 \left\{ \frac{(M_2-m_2)(M_2-m_2+R_2)}{M_2^2} \left\{ \frac{1}{r_2} - \frac{1}{R_2} \right\} + \frac{1-\theta_1}{M_2} \right\}$$

where:

R_i = the number of tags (new and old) released in year i.

r_2 = the total recoveries from release R_2 in future years, i.e. here in year 3,

z_2 = recoveries from char tagged in first year, not recovered in year 2, but recovered in year 3,

M_2 = number of tags present prior to the second years sampling, i.e. survivors of the first years release.

m_2 = number of tags recovered in second years sample.

and:

$R_i = N_i \, p_c \, p_t$

$M_2 = R_1 \, \theta_1$

$m_2 = M_2 \, p_c$

$$r_2 = R_2\ \theta_1\ p_c$$

$$z_2 = R_1\ \theta_1\ (1-p_c)\ \theta_2\ p_c$$

where:

N_i - abundance in year i

p_c - probability of capture in each years sampling, which is assumed in this analysis to be equal for all three years

p_t - proportion of fish captured that are tagged, also assumed equal for all samples

θ_2 - survival from year 2 to 3, which is assumed to be equal to θ_1.

The hypothesis that survivals are different between oiled and nonoiled areas at an alpha level of 0.05, can be tested by examining the 95% confidence intervals, if they do not overlap then the difference is significant. This exercise involved setting an expected level of abundance and survival, examining the confidence intervals achieved at different levels of tagging ($R_i = N_i\ p_c\ p_t$) and handling ($n_i = N_i\ p_c$) and choosing the sampling levels that allow us to detect a pre-defined difference in survival rates. This difference was set at 5%, and the results for three different population levels, and four levels of survival are presented in Table 2.

The tagging levels were increased by 10% to allow for sampling error in the tag recovery. The variance, and the confidence interval, depends to a great extent on m_2, r_2 and z_2, the tag recoveries in the second and third year (see equation 2). The percent of the emigration to be examined for tags that is presented in Table 2 is a minimum. The goal will be 100%, however the realized percent should not be allowed to fall below those presented in Table 2, if the objective as stated above is to be achieved.

Appendix B.

FY90 Operational Plan

Lower Kenai Peninsula Dolly Varden and Steelhead studies

FY90

OPERATIONAL PLAN

Lower Kenai Peninsula Dolly Varden and Steelhead Trout Studies

Principal Investigator: Larry Larson, Fishery Biologist II

Assisting Personnel: Thomas Balland, Fishery Biologist I
James Cofske, Fishery Technician II
R. Dederick, Fishery Technician II
Bradley Carver, Fishery Technician I
William Kline, Fishery Technician I
A. Helminski, Fishery Technician I

Date Submitted: April 8, 1989

Date Revised: May 25, 1989

APPROVED

Title	Signature	Date
Project Manager	[signature]	5/26/89
Area Manager		
Consulting Biometrician	[signature]	6-14-89
Regional Research Supervisor	[signature]	5/26/89
Regional Supervisor	[signature]	6/22/89
Biometrics Chief	[signature]	7/5/89

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SYNOPSIS

Project Title

Statistics for Selected Sport Fisheries on the Lower Kenai Peninsula, Alaska, During 1989 with Emphasis on Dolly Varden Char and Steelhead Trout.

Need

The Anchor River is the most intensively used stream by anglers on the southwestern Kenai Peninsula. While stocks of coho salmon *Oncorhynchus kisutch* and chinook salmon *O. tshawytscha* have either remained stable or increased, there are indications that the Anchor River Dolly Varden char *Salvelinus malma* and steelhead/rainbow trout *O. mykiss* populations have declined. An accurate assessment of the Dolly Varden char and steelhead/rainbow trout populations are necessary for sound resource management.

Dolly Varden char and steelhead trout life history studies on southwestern Kenai Peninsula streams are limited. Baseline data were first obtained in 1954 and 1957 and again from 1986 - 1988.

On the Anchor River, immigration of Dolly Varden char from saltwater begins about the end of June and continues throughout August. These fish are available to sport anglers throughout the summer and fall. Sport fishing regulations permit harvest of Dolly Varden char during the special chinook salmon weekend fishery in May and June, and from 1 July through 31 December.

The studies conducted in 1954 and 1957 indicated that upstream migration of adult Dolly Varden char may be only one-quarter mile per week. The first three miles immediately upstream from the mouth are the most intensely fished area and, therefore, Dolly Varden char may be subject to harvest in this area for many weeks. Although Dolly Varden char are not the target species of all anglers, the methods and means used to harvest other species are also effective for catching Dolly Varden char. By tagging Dolly Varden char in the Anchor River, future recaptures in the Anchor River and other systems will provide information which may be important to the management of this resource.

Steelhead trout immigrate into the Anchor river from saltwater beginning in mid August and continuing through October. These fish are available to sport anglers through late summer and fall. Sport fishing regulations permit hook-and-release fishing during the special chinook salmon weekend fishery in May and June, and from 1 July through 31 December. The hook-and-release policy was imposed by the Board of Fisheries for 1989 as a necessary stock conservation action. The Alaska Department of Fish and Game's assessment of steelhead trout population levels in 1988 indicated more restrictive angling regulations and continued adult population assessments are necessary to manage this resource. Future recruitment will be provided for by assuring adequate escapement levels of adult fish.

Benefits

Quantitative estimates of total harvest, daily catch rates, and age, sex, length, and maturity of the sport catch are required to develop a regulatory regime which will provide for long-term yield of the Dolly Varden resource. Also, by determining total run strength of adult fish through a weir and subtracting the total sport harvest, escapement levels can be accurately determined. The future production by these escapements will ultimately be useful in estimating optimum escapement goals. In addition, the creel survey provides timely catch and harvest information for coho salmon and catch information on steelhead trout.

Objectives

This study is designed to characterize the length and age structure of the Anchor River Dolly Varden population, estimate the contribution of Dolly Varden of unknown origin to the Anchor River recreational fishery, census the annual abundance of Anchor River Dolly Varden, and determine use of the Anchor River for Dolly Varden spawning and overwintering.

Dolly Varden:

1. Census the immigration and emigration of Dolly Varden char through a weir on the Anchor River during the period 1 July to 15 November, 1989.
2. Estimate the length frequency of immigrant and emigrant Dolly Varden at the weir by weekly intervals during the period 1 July through 15 November, 1989.
3. Estimate the sex ratio, percent spawners, age composition, and mean length-at-age of immigrant Dolly Varden, by week, at the weir during the period 1 July through 15 August, 1989.

4. To estimate the sex ratio and percent post-spawners for emigrant Dolly Varden at the weir.

5. Estimate the contribution of marked Dolly Varden to the immigrating population at the weir during the period 1 July through 15 November, 1989 and to the harvest in the Anchor River recreational fishery.

Steelhead:

6. Census the immigration of steelhead trout through a weir on the Anchor River during the period 1 July through 15 November, 1989.

7. Estimate the age and length compositions of adult steelhead trout at the weir.

Coho Salmon:

8. Census the immigration of coho salmon through a weir on the Anchor River during the period 1 July through 15 November.

9. Estimate the age and length compositions of Anchor river coho salmon.

Chinook Salmon:

10. Estimate the ordinal index of chinook salmon spawning in Deep Creek, Ninilchik River, and Anchor River.

Creel Survey:

11. Estimate the fishing effort, catch, and harvest of Dolly Varden and coho salmon; and the effort and catch of steelhead trout in the recreational fishery during the period 1 July through 15 November.

12. Estimate recreational angler demographic parameters pertaining to residency and terminal tackle preference.

Procedures

Anchor River Weir:

A weir will be installed at the upstream limit of the Anchor River intertidal zone to assess the immigration and emigration of all fish. Installation of the weir will start 1 July, 1989. The weir should be fully operational by 4 July and operate until the immigration of all adult fish have ceased entering the system (mid November).

All Dolly Varden immigrants will be counted and observed for tags (Floy FD-67 Anchor Tags). In addition, Dolly Varden immigrants will be sampled for age, sex, fork length, and relative maturity. Gonad development will be used to determine relative maturity of each fish. All emigrating Dolly Varden will be sampled for length and all fish over 200 mm in fork length will be tagged with Floy FD-67 Anchor Tags. All post-spawner Dolly Varden will be sexed. Post spawners will be identified by external characteristics. Any Dolly Varden mortalities will be sampled for age (removing both otoliths), fork length, and relative maturity.

Age and length composition of adult coho salmon and steelhead trout will be estimated. Age will be determined from scale analysis.

Anchor River Creel Survey:

A stratified random creel survey will be conducted to estimate recreational angler effort, harvest, and catch of Dolly Varden char and coho salmon. In addition, recreational angler effort and catch of steelhead trout will be estimated. Angler counts will be made to estimate effort in angler-hours and interviews will be conducted to estimate catch and harvest rates.

Each day is stratified into two sample periods; the number of hours in each period varying from month to month (dependant on available daylight). Count hours are randomly selected within these periods. Two counts will be conducted daily, with a minimum of four hours between counts. Angler counts will be conducted from saltwater to one-quarter mile above the junction of the

North and South forks on the South Fork, Blackwater Bend, and the South Fork Wayside. Counts downstream and upstream of the weir will be kept separate during the operation of the weir.

Completed-trip angler interviews will be conducted around the randomly selected count hours. Interviews from anglers who fished upstream or downstream from the weir will be kept separate. Creel survey personnel will interview any angler exiting the stream bank whether the angler is returning to a campsite, vehicle, or exiting the fishery. Information collected will include, but not be limited to: harvest by species, time spent fishing, presence or absence of a tag on fish caught, residency, and terminal tackle. Biological sampling (age, sex, and length data) of Dolly Varden char and coho salmon will be obtained at every available opportunity.

Spawning Ground Sampling:

The ordinal index of spawning chinook salmon in Deep Creek, Ninilchik and Anchor Rivers will be estimated using aerial and ground counts of live and dead salmon. One survey will be conducted during the last week of July.

Location

The Anchor River weir will be located adjacent to the Alaska Department of Fish and Game (ADF&G) cabin. This site is approximately 1.5km upstream from saltwater and is in the vicinity of the "Dudas Hole", a local name for a popular fishing location.

The Anchor River creel survey will be conducted from saltwater upstream approximately 3.75km, to include approximately 0.4km above the junction of the North and South forks on the South fork, and approximately 0.4km from Blackwater Bend upstream to the South Fork Wayside.

Deliverables

The results of this project will be presented in an Annual Report of Performance, Federal Aid to Fish Restoration, as part of the Alaska Department of Fish and Game's Fisheries Data Series.

Budget Summary

Projected FY90 Costs:

Line Item	Category	Cost ($K)
100	Personnel Services	118.6
200	Travel	3.3
300	Contractual	11.1
400	Commodities	5.3
500	Equipment	5.1
	Total	143.4

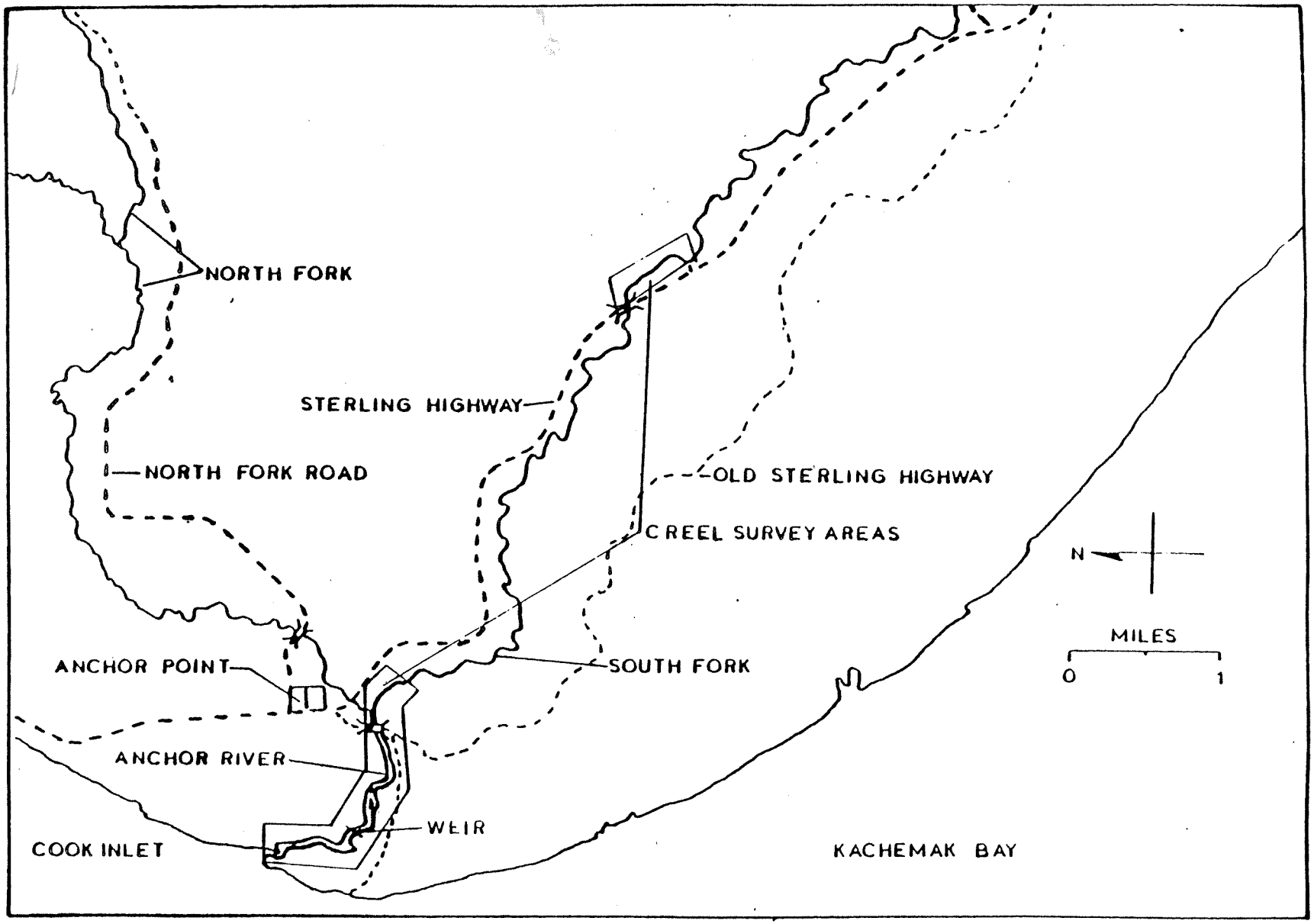
Budget Manager: Larry Larson
Project Personnel:

PCN	Name	Level	Months
4017	Larry Larson	FB II	12.0
4162	Tom Balland	FB I	5.5
4144	James Cofske	FT II	4.5
4172	Bill Kline	FT I	4.5
4211	(Vacant)	FT I	4.5

INTRODUCTION

The Anchor and Ninilchik rivers and Deep Creek on the lower Kenai Peninsula (Figure 1) support recreational fisheries for chinook salmon *Oncorhynchus tshawytscha*, coho salmon *O. kisutch*, and pink salmon *O. gorbuscha*; Dolly Varden char *Salvelinus malma*; and anadromous (steelhead trout) and resident rainbow trout *O. mykiss*. The downstream sections of each of these streams are crossed by the Sterling Highway making them easily accessible to the fishing public. Much of the river frontage on these streams is publicly owned, providing ample camping and parking areas. Due to their relatively small size, all fishing in these streams is conducted from the bank. Of the three streams, the Anchor River is the most heavily used providing an average of 33,831 recreational fishing days (angler-days) annually from 1977 through 1987 (Mills 1979-1988). The Ninilchik River and Deep Creek provided an average of 12,898 and 13,819 angler-days, respectively, over this same period.

The fisheries targeting chinook salmon, coho salmon, steelhead trout, and Dolly Varden char are of major importance to recreational anglers on the lower Kenai Peninsula, whereas the fisheries targeting resident rainbow trout and pink salmon are of lesser importance. The recreational fishery for Dolly Varden char in the Anchor River is one of the largest in Alaska and is of particular concern to resource managers. During the period 1977 to 1983, the harvest from this fishery averaged nearly 15,000 fish annually (Mills 1979-1984). In 1984, regulations for this fishery became more restrictive as the bag and possession limits were reduced from ten fish to five and the use of bait was prohibited after 16 September. Since these regulations have been in effect, the harvest of Dolly Varden char has averaged approximately 5,000 fish


NORTH FORK
STERLING HIGHWAY
NORTH FORK ROAD
OLD STERLING HIGHWAY
CREEL SURVEY AREAS
N
MILES
0
1
ANCHOR POINT
SOUTH FORK
ANCHOR RIVER
WEIR
COOK INLET
KACHEMAK BAY

(Mills 1985-1988). Although a marked decline has been observed in the harvest of Dolly Varden char after initiation of the new regulations, concern has been expressed that the decline may also reflect a depressed population. To date, the Department has conducted quantitative assessments of this resource from 1987-1988 and has estimated angler harvest in the statewide mail survey (Mills 1979-1988).

The Department initiated a pilot program in 1986 to investigate the Dolly Varden char resource of the lower Kenai Peninsula streams. The program involved angler interviews to estimate catch and harvest rates, aerial surveys to estimate angler distribution, and tagging fish to determine migrational patterns among lower Kenai Peninsula streams (Nelson et al. 1987). The program was expanded in 1987 and 1988 to include more detailed estimates of effort and harvest in the Anchor River fishery, census of the Dolly Varden char immigration into and emigration out of the Anchor River, a more aggressive tagging program in the three major streams, and estimation of length and age parameters. The long-term goal of the program is to estimate sustainable yield for this resource.

Information pertaining to the Dolly Varden char, steelhead/rainbow trout, and coho salmon fisheries has been presented by Allin (1954, 1957), Balland (1985-1986), Nelson et al. (1987), Larson et al. (1988), Wallis and Balland (1981-1984) and Wallis and Hammarstrom (1979-1982). Harvest and effort estimates have been reported by Mills (1979-1988).

OBJECTIVES

Dolly Varden

1. Census the immigration and emigration of Dolly Varden char through a weir on the Anchor River during the periods 1 July to 15 November 1989.

2. Estimate the length frequency of immigrant and emigrant Dolly Varden at the weir by weekly intervals during the period 1 July through 15 November, such that the estimated proportion by length class is within ±10% of the true value 90% of the time.

3. Estimate the sex ratio, percent spawners, age composition and mean length-at-age of immigrant Dolly Varden by week at the weir during the period 1 July through 15 August, such that the estimates are within ±10% of the true value 90% of the time.

4. Estimate percent post-spawners for emigrant Dolly Varden at the weir and estimate the sex ratio for the post-spawners, such that the estimated proportion by class is within ±10% of the true value 90% of the time.

5. Estimate the contribution of marked Dolly Varden to the immigrating population at the weir during the period 1 July through 15 November and to the harvest in the Anchor River recreational fishery, such that the estimate is within ±10% of the true value 80% of the time.

Steelhead

6. Census the immigration of steelhead trout through a weir on the Anchor river during the period 1 July through 15 November.

7. Estimate the age and length composition of adult steelhead trout at the weir, such that the estimated proportion by age and length class is within ±10% of the true value 90% of the time.

Coho Salmon

8. Census the immigration of coho salmon through a weir on the Anchor River during the period 1 July through 15 November.

9. Estimate the age and length composition of Anchor river coho salmon, such that the estimated proportion by age and length class is within ±10% of the true value 90% of the time.

Chinook Salmon

10. Estimate the ordinal index of chinook salmon spawning in Deep Creek, Ninilchik River, and Anchor River.

Creel Survey

11. Estimate the fishing effort, catch, and harvest of Dolly Varden and coho salmon; and the effort and catch of steelhead trout in the recreational fishery during the period 1 July through 15 November, such that the estimated effort is within ±10% of the true value 95% of the time, and the estimated harvest and catch are within ±15% of the true values 95% of the time,

12. Estimate recreational angler demographic parameters pertaining to residency and terminal tackle preference.

PROJECT PROCEDURES

Study Design

This is the third year of what is envisioned to be a long-term study of the lower Kenai Peninsula Dolly Varden char resource. Dynamics of large populations of anadromous Dolly Varden (Armstrong 1965) or Arctic char (McBride 1980) are often complex because they typically exhibit complicated migratory and homing patterns and it is necessary to determine the stock structure of Dolly Varden char which return to lower Kenai Peninsula streams. The Anchor River, which is the major study stream, is a non-lake system and following Armstrong's model (1974) the Dolly Varden from this system, upon leaving as smolt in the spring, will overwinter in a different lake system, and only return as spawners. The assumption is therefore made that the immigrating Dolly Varden consist of returning Anchor River spawners and non-spawners of unknown origin and that these will all emigrate the summer and fall of the same year to enter an overwintering lake.

This study will provide information to confirm this model by censusing the summer and fall immigrants and emigrants and estimating the percent of these that are spawning fish. Acquisition of basic population data such as a total census, harvest, length and age composition and relative maturity, and exploitation and contribution rates to the fishery of Anchor River and non-Anchor River fish, will provide the means to estimate key population parameters and allow estimation of maximum sustained yield (MSY). Since this fishery is complicated by concurrent fisheries for other species, it is also necessary to acquire specific fisheries information on all species so that additional regulatory measures (if necessary) can be effectively implemented.

Abundance of immigrating and emigrating Dolly Varden char will be censused through the Anchor River weir (Objective 1). The immigration of Dolly Varden will be sampled randomly at the rate of 130 fish per week from 1 July through 15 November. These samples will be used to estimate Dolly Varden length frequency (Objective 2), sex ratios, relative maturity and percent spawners, and age frequency and mean length at age (Objectives 3 and 4). All emigrant Dolly Varden will be measured for fork length and all post spawners will be identified and sexed (Objective 4). All emigrants over 200mm will be tagged with a Floy FD-67 Anchor Tag (Objective 5) during the periods 1 July through 15 November.

Dolly Varden have been tagged for the past two years. So during the immigration, as many Dolly Varden as possible will be examined for tags. If subsampling is necessary the number examined and recaptured will be recorded

as well as number passed without examination. All immigrant fish with tags will be measured for length. During emigration all fish will be examined for tags.

Abundance of steelhead trout adults immigrating through the Anchor River weir will be censused 1 July to 15 November (Objective 6). For this period 130 steelhead will be sampled to estimate age and length compositions (Objective 7).

Abundance of coho salmon adults immigrating through the Anchor River weir will be censused 1 July to 15 November (Objective 8). During this period 130 coho salmon will be sampled randomly to estimate age and length composition at the weir and in the creel (Objective 9).

The chinook salmon ordinal index will be estimated for the Anchor River, Deep Creek, and Ninilchik River during the last week of July (Objective 10). Both a ground and an aerial survey will be conducted in an index area of each stream. Additional aerial surveys will be conducted on the remaining spawning areas of each stream. The ratio of the ground survey/the aerial survey for the index site will be used to expand all aerial surveys from the remaining spawning areas. The expansion factor will not be used if the count from the ground survey at the index site is less than the count from the aerial survey.

Anchor River Creel Survey:

A roving creel survey (Neuhold and Lu, 1957) will be conducted on the Anchor River immediately after the Anchor River weir is operational. Sampling is

stratified by weekdays and weekend/holidays and the river is also stratified into two areas upstream and downstream of the weir. The length of a fishing day varies with the amount of available daylight and time of the year. During July and August, the fishing day is 16 hours (0600-2200 hours); September, 12 hours (0800-2000 hours); and October through 15 November, 8 hours (1000-1800 hours). Each sample day is stratified into two equal periods (A and B). Two sampling times, each 4.0 hours in duration, are selected randomly from each period on weekend/ holidays and on three randomly selected weekdays each week.

Sport fishing effort is estimated in units of angler-hours using a stratified random sample design. Angler counts take approximately one hour to complete and are considered to be instantaneous. Catch rates (number of fish harvested and released per hour) for each species are estimated from completed-trip angler interviews only. The estimated number of fish caught by the fishery is the product of the effort and catch rate estimates. Harvest (fish kept) is computed similarly using effort and harvest rate estimates (Objective 11).

One angler count is made during each interview period. A starting time is randomly selected in each count period subject to the constraint that there are at least four hours between counts. This is done to minimize the covariance between counts in adjacent periods. Only anglers actively engaged in fishing during the count hour will be counted. Angler counts will be recorded separately for the area downstream and upstream of the weir during weir operations. The interview will also record hours fished and fish caught and released separately for the two areas. Only completed interviews are used.

Data Collection

Anchor River Weir:

A weir will be installed approximately 1.5km upstream from the saltwater terminus of the Anchor River. The weir will be operated by a three-person crew billeted in a cabin located adjacent to the weir. The weir pickets are 1.25 cm diameter solid aluminum rods placed in an aluminum channel framework with a 1.25 cm gap between pickets. The channel frames are 3.6 meters long and 1.05 meters high. The aluminum frames rest against wooden tripods spaced approximately three meters apart. Traps will be installed to capture both up- and downstream migrating fish.

The weir will be modified by incorporating a sixty foot floating weir design in the main river channel section of the weir. The floating weir structure will consist of 15 panels, each 4.5 meters long and 1.6 meters wide. The pickets of each panel will consist of 18.75 mm, schedule 40, PVC pipe spaced 12.5 mm apart. The construction and installation of the floating weir will be accomplished by two non-permanent employees during the period 1 July through 15 August.

Weir activities will be recorded in a daily log. The date, activity, start time and stop time will be recorded. Immigrating and emigrating Dolly Varden will be detained in the forward holding area of upstream and downstream traps. All fish in the upstream trap will be examined for tags and all immigrating tagged fish will be measured for length. The number of recaptures, the number without tags, and those that are dead will be recorded on the weir count form (Appendix A3) by trap load. If all immigrants cannot be examined due to sheer

numbers, then the remaining fish will be counted with tally wackers and the counts recorded separately on the weir count form. Other species will also be recorded.

All emigrating Dolly Varden will be examined for tags in the downstream trap. All fish will be measured and those over 200mm tagged. Post spawners will be identified by external characteristics, i.e., coloration and sexed (kype on males). Data will be recorded by trap load on weir count forms and data will be transferred as soon as possible to LOTUS worksheets. Information recorded will include direction of travel (upstream/downstream), number per species, number of tagged Dolly Varden recaptured and number of new tags released, if any. Length, sex, and tag number information will be recorded on tagging-length mark-sense forms or on data recorders.

Paired otolith and fork length (measured from tip-of-snout to fork-of-tail to the nearest millimeter) data to estimate age-length relationships for Dolly Varden will be collected at the rate of 130 samples per week from 1 July through 15 August. Sex and relative maturity will also be recorded for these fish. These samples will be chosen randomly by selecting a trap load and sampling all fish from that trap load. In addition, all Dolly Varden mortalities recovered at the weir will be sampled for length, age, sex and relative maturity. Both otoliths and gonads will be removed from each Dolly Varden sampled and stored in an envelope for future age and maturity determination. Information on each envelope will include: date, species, sex, source (weir, creel, or mortality), fork length, cause of death (if known),

age and maturity state These data will be recorded on mark-sense forms upon completion of age and maturity determination.

Relative maturity of each female Dolly Varden will be determined by using the criteria described by Blackett (1968):

State I. Immature female: completely undeveloped ovary, eggs minute (usually less than 0.90 mm in diameter) and yolkless.

State II. Maturing female: maturing ovary will develop by spawning period, eggs usually larger than 1.75 mm in diameter and appear to be approaching an advanced stage of maturity. Oil droplets are present in the eggs and vessel structure is well developed in the ovarian tissue.

State III. Completely mature female: ovaries have reached a degree of maturity allowing the eggs to be easily stripped from the fish with only slight pressure.

State IV. Completely spawned female: only vestiges of recently spawned eggs in the ovary; i.e., ovary appears as a string with many minute recruitment eggs embedded in the tissue.

State V. Immature female but shows a degree of development: Ovaries do not appear as if they would mature this year but development is definitely more advanced than State I. Egg diameters are usually greater than 0.90 mm but less than 1.75 mm. Ovary size is large enough to indicate spawning next year.

One or more trips will be made to English Bay Lakes in the spring and fall of 1989 and 1990. Dolly Varden will be sampled at the outlet of the most downstream lake of the drainage with a weir. All Dolly Varden greater than 200 mm in fork length captured will be anesthetized with CO_2, measured for fork length, and tagged with Floy FD-67 Anchor Tags. Recovery of these tagged fish will occur at the Anchor River weir during 1990.

Steelhead trout adults immigrating through the Anchor River weir will be censused and recorded as described above. Age (removing three scales from the

preferred area) and fork length will be measured for a random sample of 130 fish.

Coho salmon adults immigrating through the Anchor River weir will be censused and recorded as described above. Age (removing three scales from the preferred area) and mid-eye to fork length will be collected for 130 salmon during the period 1 July through 15 November.

The chinook salmon ordinal index for the Anchor river, Deep Creek, and Ninilchik River will be conducted the last week in July. Aerial and ground survey personnel will keep observed counts of live and dead chinook salmon separate. Data will be entered at the end of the survey in a LOTUS worksheet file.

Additional weir duties which will be recorded in a daily log include:

1) Cleaning debris from the weir. The time and date of this action will be noted.

2) Water temperature and depth measurements will be obtained daily at 2200 hours. The station for these measurements will be determined by the project leader after the weir has been installed.

Anchor River Creel Survey:

One Fishery Technician II will conduct the creel survey from 1 July through 15 November. At the designated time on the schedule (Appendix Table A1), the survey person will either begin conducting interviews or initiate an instantaneous angler count from one end of the survey area. Every effort should be made to complete the count in less than one hour. During the instantaneous count, the survey person will count only anglers actively

engaged in fishing. Anglers counted upstream and downstream of the weir will be recorded separately. Count data will be entered on mark-sense forms.

Completed-trip angler interviews will be conducted at campgrounds and key access points. If an angler intends to continue fishing that same day, creel personnel will indicate incomplete-trip on the mark-sense form. Completed-trip angler interviews will be collected both from anglers who claim they are through fishing for the day or have fished the day before on the Anchor River. The previous day interview is inclusive of all hours fished and all fish captured, to include any incomplete-trip interview information which may have already been collected the day before. Previous day interviews from anglers who were interviewed as a completed-trip angler the previous day will not be collected. Any anglers who fished both upstream and downstream of the weir location will be interviewed twice: once for the period of time he/she fished upstream and again for the period of time he/she fished downstream of the weir. This will allow harvest and catch estimates to be made in both river areas. Demographic information will only be recorded once for each completed angler fishing day.

The following information will be obtained from each angler interviewed: number of hours fished, number of fish harvested (by species), number of fish released (by species), target species, terminal gear (through 15 August only): bait, artificial, or both; and residence.

Data will be recorded on mark-sense forms. The option fields of the mark-sense forms will be used to record fishing location, date of interview, and

demographic information. Option field 1 will record the amount of angler fishing time expended upstream (indicated by a "0") or downstream (indicated by a "1") of the weir. Option field 2 will record whether a completed angler interview occurred on the same day (indicated by a "0") or the previous day (indicated by a "1"). Option field 3 will record whether an angler used bait (indicated by a "1"), an artificial lure (indicated by a "2"), or both bait and lures (indicated by a "3").

Biological sampling of Dolly Varden char will be conducted to estimate sex, length-frequency, and age of the harvest. Tip-of-snout to fork-of-tail length will be measured to the nearest mm on each fished sampled. Otoliths and gonads will be removed whenever possible, otoliths stored in envelopes and gonads in plastic bags. All samples will be clearly marked with labels. A total of 130 fish per month will be sampled during July, August and October. All data will be recorded on mark-sense forms by crew leader.

Coho salmon will be sampled to estimate age, sex, and length composition. a total of 130 fish will be sampled for three scales from the preferred area (on a diagonal line from the posterior portion of the dorsal fin to the anterior portion of the anal fin and three rows above the lateral line), sex, and length (mid-eye to fork-of-tail). This sample size was selected to provide desired levels of accuracy and precision, given 15% unreadable scales (Thompson 1987). Data will be recorded on mark-sense forms and scales stored on adhesive-coated cards.

Data Reduction

Weir counts will be entered into a LOTUS 1-2-3 spreadsheet. Angler count and interview mark-sense forms will be processed monthly. Dates for previous day interviews will be adjusted to the correct (previous day's) date using program RECODANC. Sublocations will also be coded to either above or below the weir, based on what is entered in the option 1 field, using RECODANC.

Otoliths and gonads will be processed, otoliths aged and maturity state of gonad determined at the Anchor River cabin. Upon completion all data will be recorded on mark-sense forms. All mark-sense forms will be sent to RTS in Anchorage at least twice a month for processing.

Data Analysis

Total weir counts of immigrants and emigrants will be tabulated by species at the end of the season. Age and length-frequencies will be constructed for each sample group of Dolly Varden. These groups include spawners and non-spawners at the weir by week for immigrants and emigrants, and for spawners and non-spawners in the creel. The lengths of all fish measured by week will be compared with a one way ANOVA to determine if the size of the fish changes over time. Chi-square tests will be used to compare the number of fish in each age group for each week to determine if age composition changes over time. Chi-square tests will also be used to compare the number of spawners and nonspawners present each to determine if the proportion of spawners changes over time. To test for a difference in mean length at age between spawners and nonspawners, a series of T tests will be used, comparing the mean length at each age.

The contribution of tagged populations to the Anchor River weir and fishery will by estimated by,

$$\hat{n}_{ij} = \frac{m_{ij} \; n_{2j}}{h \; N}$$ Clark and Bernard (1987)

where,

$\hat{n}_{ij}$ = estimated total number of fish in area j from population i

m_{ij} = number of tags from population i observed in area j

n_{2j} = number of fish examined for tags in area j

N = total number of fish in area j

h = proportion of fish tagged in population i.

and the variance of $\hat{n}_{ij}$ is approximately estimated by,

$$Var(\hat{n}_{ij}) = \frac{m_{ij}}{\ddot{A}^2 \; h^2}(1 - \phi\theta)$$

where,

ϕ = proportion examined for tags = $\frac{n_2}{N}$

Effort (in angler-hours) will be estimated using a stratified random sample design with periods as separate strata. Estimates for months and river reach are considered independent and seasonal estimates of effort and variance are the sum of these quantities. LOTUS 1-2-3 worksheets ROVING 2 (Conrad 1987) will be used to estimate effort and its variance.

Angler harvest rates (HPUE) will be estimated from completed-trip interview data. The mean effort and harvest per angler will be estimated for each species, month and river reach. The assumption is made that there is no

difference between periods within days in harvest rates. The variance of these means will be estimated using a two-stage design with days as the primary sample units and anglers as the secondary sample units (Von Geldern and Tomlinson 1973). Harvest per angler-hour will be estimated as the quotient of the mean harvest and mean effort estimates. The variance of the harvest rate will be estimated using the approximation for the quotient of the means of two random variables (Jessen 1978). Program AISUM89 will be used to generate these estimates.

Angler catch rates (CPUE) will be estimated similarly to angler harvest rates (HPUE) but will include all fish (by species) caught (retained and released).

Total harvest and catch will be estimated as the product of the angler effort and angler catch rate estimates. The variance of these quantities will be estimated following the procedure for the product of two independent random variables (Goodman 1960).

SCHEDULES

The creel survey schedule for 1988 is presented in Appendix A1.

Task Target Dates:

1. Prepare equipment for the field season.	June 12-June 30	(Balland)
2. Install Anchor River weir.	July 1-July 4	(Larson) (Balland) (Carver) (Kline)
3. Construct floating weir	July 1- Aug 15	(non-perm) (non-perm)
4. Operate Anchor River weir.	July 4-Nov 15	(Larson) (Balland)

		(Carver) (Kline)
5. Anchor River creel survey.	July 1-Nov 15	(Cofske)
6. Weir data collection, computer entry, and analysis.	daily	(Larson) (Balland)
7. Otolith and scale aging.	daily	(Balland)
8. Initial harvest and effort estimates	monthly	(Larson)
9. Weir counts and preliminary harvest and effort estimates (DV,SS,SH) for Management Report	Oct 1	(Larson)
10. Scales/otoliths read (from creel, DC, & NR) and final analysis completed	Dec 15	(Balland)
11. Final creel estimates.	Dec 15	(Larson)
12. Detailed data analysis.	Jan 15	(Larson)
13. Annual Report (draft) submitted.	Feb 13	(Larson) (editing Marianna)
14. FY91 Operation Plan Synopsis	Mar 15	(Larson)
15. FY91 Operation Plan.	April 30	(Larson)

REPORTING

A complete report will be prepared in accordance with the requirements of the Federal Aid contract and included as part of the S-32 Annual Report of Progress. This report will be published as a Division of Sport Fisheries Fishery Data Series Report.

RESPONSIBILITIES

Thomas Balland, Fishery Biologist I, 7/1 thru 12/15 and 4/16 thru 6/30. Tom Balland's primary responsibility will be the operation of a weir on the Anchor River. He will assure all data collected is accurately recorded on the

appropriate forms and/or computer files. All age/length data collected from this project (weir, creel, and seining) will be summarized by 15 Dec. In addition, Balland will be crew leader for two Fish and Wildlife Technician I's assigned to the weir, and one FT II assigned to creel survey. Balland will collect and review for accuracy all creel data prior to presenting it monthly to the project leader, Larry Larson. Balland will be responsible for maintaining a State vehicle and two cabins located on the Anchor River. Any purchases will be processed immediately by forwarding all invoices and Field Purchase Orders to the project leader. Weekly reports (submitted every two weeks) and monthly time sheets (submitted no later than the 12th of each month) will be completed and submitted to Larry Larson, to include those completed by Balland's assigned technicians.

From 16 April - 1 May, Balland will assist Larry Larson in the preparation of the Anchor River weir equipment and housing facilities for the spring field season. Any weir repairs, modifications, or logistical needs and improvements to the housing facilities will be accomplished prior to 1 May. From 1 May - 30 June Balland will be responsible for the operation of the weir and supervise two Fish and Wildlife Technicians as described above.

Bradley Carver, Fisheries Technician I, 7/1 thru 11/15.
William Kline, Fisheries Technician I, 7/1 thru 11/15.

These technicians will assist Thomas Balland on the Anchor River Weir. They will assist in the assembly and disassembly of a fish weir. They will maintain the weir structure under the immediate supervision of either the

project leader or crew leader. They will also maintain a log of all maintenance activities.

They will collect biological samples to include data on species, size, age, and sex composition; perform fish tagging, scale mounting, and recording of water depth and water temperature. Identify/count adult and smolt species of fish, collect scales, otoliths, ovaries and biological samples. All data collected will either be entered on mark-sense forms or a computer systems. Basic daily summaries of fish counts will be computed on computer spreadsheets.

James Cofske, Fisheries Technician II, 7/1 thru 11/15

Cofske's primary responsibility is to perform the creel survey on the Anchor River, however, he may assist in the installation of the weir until July 4. His creel duties include following the schedule as closely as possible, conducting the angler counts, interviews, and collecting the biological data indicated. All data collected daily will be submitted to Thomas Balland.

BUDGET SUMMARY

A detailed breakdown of the FY90 budget is in Appendix A4.

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APPENDIX A1

Anchor River creel survey schedule

Appendix Table A1. Anchor River creel survey schedule from July through 15 November, 1989.

Date	Wknd	Start	Stop	Start	Stop	Ct 1	Ct 2
01-Jul							
02-Jul							
03-Jul		1000	1400	1800	2200	1000	2100
04-Jul	*	0700	1100	1600	2000	0700	1600
05-Jul		OFF					
06-Jul		OFF					
07-Jul		0800	1200	1700	2100	0800	2000
08-Jul	*	0600	1000	1600	2000	0800	1800
09-Jul	*	0700	1100	1600	2000	0800	1600
10-Jul		0700	1100	1500	1900	1000	1500
11-Jul		0600	1000	1800	2200	0900	2100
12-Jul		0700	1100	1400	1800	0900	1500
13-Jul		OFF					
14-Jul		OFF					
15-Jul	*	0600	1000	1700	2100	0600	1800
16-Jul	*	0800	1200	1800	2200	0900	1800
17-Jul		OFF					
18-Jul		OFF					
19-Jul		0600	1000	1500	1900	0800	1800
20-Jul		0900	1300	1600	2000	1100	1600
21-Jul		0700	1100	1600	2000	0700	1800
22-Jul	*	0700	1100	1700	2100	0800	1900
23-Jul	*	0900	1300	1700	2100	1100	1800
24-Jul		0600	1000	1600	2000	0700	1700
25-Jul		1000	1400	1800	2200	1400	2000
26-Jul		OFF					
27-Jul		OFF					
28-Jul		1000	1400	1800	2200	1200	1900
29-Jul	*	0600	1000	1400	1800	0600	1400
30-Jul	*	0900	1300	1400	1800	1100	1500
31-Jul		0700	1100	1500	1900	0700	1600

-CONTINUED-

Appendix Table A1. Anchor River creel survey schedule from July through 15 November, 1989 (continued).

Date	Wknd	Start	Stop	Start	Stop	Ct 1	Ct 2
01-Aug		OFF					
02-Aug		OFF					
03-Aug		1000	1400	1800	2200	1000	2000
04-Aug		0700	1100	1600	2000	0900	1700
05-Aug	*	0800	1200	1700	2100	1100	1700
06-Aug	*	0600	1000	1600	2000	0900	1800
07-Aug		OFF					
08-Aug		OFF					
09-Aug		0700	1100	1500	1900	0900	1500
10-Aug		0600	1000	1800	2200	0800	2000
11-Aug		0700	1100	1400	1800	0800	1700
12-Aug	*	0600	1000	1700	2100	0600	1900
13-Aug	*	0800	1000	1800	2200	0900	1800
14-Aug		0600	1000	1500	1900	0900	1700
15-Aug		0900	1300	1600	2000	1000	1900
16-Aug		0700	1000	1700	2100	0800	2000
17-Aug		OFF					
18-Aug		OFF					
19-Aug	*	0700	1100	1600	2000	1000	1800
20-Aug	*	0900	1300	1700	2100	1200	2000
21-Aug		0600	1000	1600	2000	0800	1800
22-Aug		1000	1400	1800	2200	1300	1800
23-Aug		OFF					
24-Aug		OFF					
25-Aug		1000	1400	1600	2000	1100	1600
26-Aug		0600	1000	1400	1800	0800	1500
27-Aug	*	1000	1400	1400	1800	1200	1500
28-Aug	*	0900	1300	1500	1900	0900	1700
29-Aug		0700	1100	1400	1800	1000	1700
30-Aug		OFF					
31-Aug		OFF					

-CONTINUED-

Appendix Table A1. Anchor River creel survey schedule from July through 15 November, 1989 (continued).

Date	Wknd	Start	Stop	Start	Stop	Ct 1	Ct 2
01-Sep		1000	1400	1600	2000	1300	1700
02-Sep	*	1000	1400	1500	1900	1000	1700
03-Sep	*	1000	1400	1600	2000	1100	1700
04-Sep	*	0900	1300	1500	1900	0900	1500
05-Sep		0800	1200	1600	2000	0800	1900
06-Sep		1000	1400	1500	1900	1000	1500
07-Sep		OFF					
08-Sep		OFF					
09-Sep	*	0900	1300	1600	2000	0900	1900
10-Sep	*	0900	1300	1500	1900	1000	1500
11-Sep		OFF					
12-Sep		OFF					
13-Sep		0800	1200	1600	2000	1100	1900
14-Sep		0800	1200	1400	1800	1100	1500
15-Sep		0800	1200	1500	1900	0900	1700
16-Sep	*	0900	1300	1500	1900	1200	1700
17-Sep	*	1000	1400	1600	2000	1200	1800
18-Sep		0800	1200	1600	2000	0800	1900
19-Sep		0900	1300	1500	1900	1200	1700
20-Sep		1000	1400	1600	2000	1000	1900
21-Sep		OFF					
22-Sep		OFF					
23-Sep	*	0900	1300	1600	2000	1000	1600
24-Sep	*	0800	1200	1400	1800	1100	1700
25-Sep		1000	1400	1500	1900	1200	1500
26-Sep		0800	1200	1500	1900	0800	1600
27-Sep		OFF					
28-Sep		OFF					
29-Sep		0800	1200	1400	1800	0900	1400
30-Sep	*	1000	1400	1500	1900	1300	1700

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Appendix Table A1. Anchor River creel survey schedule from July through 15 November, 1989 (continued).

Date	Wknd	Start	Stop	Start	Stop	Ct 1	Ct 2	
01-Oct	*	1000	1400	1400	1800	1100	1600	
02-Oct		1000	1400	1400	1800	1000	1500	
03-Oct		OFF						
04-Oct		OFF						
05-Oct		1000	1400	1400	1800	1300	1700	
06-Oct		1000	1400	1400	1800	1200	1700	
07-Oct	*	1000	1400	1400	1800	1100	1700	
08-Oct	*	1000	1400	1400	1800	1100	1500	
09-Oct		1000	1400	1400	1800	1100	1700	
10-Oct		1000	1400	1400	1800	1300	1700	
11-Oct		OFF						
12-Oct		OFF						
13-Oct		1000	1400	1400	1800	1200	1700	
14-Oct	*	1000	1400	1400	1800	1000	1700	
15-Oct	*	1000	1400	1400	1800	1000	1400	
16-Oct		1000	1400	1400	1800	1300	1700	
17-Oct		1000	1400	1400	1800	1300	1700	
18-Oct	*	1000	1400	1400	1800	1200	1600	
19-Oct		OFF						
20-Oct		OFF						
21-Oct	*	1000	1400	1400	1800	1100	1600	
22-Oct	*	1000	1400	1400	1800	1000	1700	
23-Oct		1000	1400	1400	1800	1300	1700	
24-Oct		OFF						
25-Oct		OFF						
26-Oct		1000	1400	1400	1800	1100	1600	
27-Oct		1000	1400	1400	1800	1100	1600	
28-Oct	*	1000	1400	1400	1800	1300	1700	END DST
29-Oct	*	0900	1300	1300	1700	1100	1600	
30-Oct		0900	1300	1300	1700	1100	1500	
31-Oct		OFF						

-CONTINUED-

Appendix Table A1. Anchor River creel survey schedule from July through 15 November, 1989 (continued).

Date	Wknd	Start	Stop	Start	Stop	Ct 1	Ct 2
01-Nov		OFF					
02-Nov		0900	1300	1300	1700	1000	1600
03-Nov		0900	1300	1300	1700	0900	1300
04-Nov	*	0900	1300	1300	1700	1100	1500
05-Nov	*	0900	1300	1300	1700	1000	1400
06-Nov		0900	1300	1300	1700	1100	1600
07-Nov		0900	1300	1300	1700	1000	1400
08-Nov		OFF					
09-Nov		OFF					
10-Nov		0900	1300	1300	1700	1000	1600
11-Nov	*	0900	1300	1300	1700	1200	1600
12-Nov	*	0900	1300	1300	1700	0900	1500
13-Nov		0900	1300	1300	1700	0900	1400
14-Nov		0900	1300	1300	1700	1100	1500

APPENDIX A2

Anchor River weir schedule

Appendix Table A2. Anchor River Weir schedule from July through 15 November, 1989.

Date	Wknd	Balland	Carver	Kline	Non-perms
01-Jul	*	INSTALL WEIR			
02-Jul	*	INSTALL WEIR			
03-Jul		INSTALL WEIR			
04-Jul	*	OFF	0000-2400	2200-2400	OFF
05-Jul		OFF	0000-0530	0000-2400	0800-1630
06-Jul		2200-2400	OFF	0000-2400	0800-1630
07-Jul		0000-2400	OFF	0000-2400	0800-1630
08-Jul	*	0000-2400	2200-2400	0000-0530	OFF
09-Jul	*	0000-2400	0000-2400	OFF	OFF
10-Jul		0000-0530	0000-2400	OFF	0800-1630
11-Jul		OFF	0000-2400	2200-2400	0800-1630
12-Jul		OFF	0000-0530	0000-2400	0800-1630
13-Jul		2200-2400	OFF	0000-2400	0800-1630
14-Jul		0000-2400	OFF	0000-2400	0800-1630
15-Jul	*	0000-2400	2200-2400	0000-0530	OFF
16-Jul	*	0000-2400	0000-2400	OFF	OFF
17-Jul		0000-0530	0000-2400	OFF	0800-1630
18-Jul		OFF	0000-2400	2200-2400	0800-1630
19-Jul		OFF	0000-0530	0000-2400	0800-1630
20-Jul		2200-2400	OFF	0000-2400	0800-1630
21-Jul		0000-2400	OFF	0000-2400	0800-1630
22-Jul	*	0000-2400	2200-2400	0000-0530	OFF
23-Jul	*	0000-2400	0000-2400	OFF	OFF
24-Jul		0000-0530	0000-2400	OFF	0800-1630
25-Jul		OFF	0000-2400	2200-2400	0800-1630
26-Jul		OFF	0000-0530	0000-2400	0800-1630
27-Jul		2200-2400	OFF	0000-2400	0800-1630
28-Jul		0000-2400	OFF	0000-2400	0800-1630
29-Jul	*	0000-2400	2200-2400	0000-0530	OFF
30-Jul	*	0000-2400	0000-2400	OFF	OFF
31-Jul		0000-0530	0000-2400	OFF	0800-1630

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Appendix Table A2. Anchor River Weir schedule from July through 15 November, 1989 (continued).

Date	Wknd	Balland	Carver	Kline	Non-perms
01-Aug		OFF	0000-2400	2200-2400	0800-1630
02-Aug		OFF	0000-0530	0000-2400	0800-1630
03-Aug		2200-2400	OFF	0000-2400	0800-1630
04-Aug		0000-2400	OFF	0000-2400	0800-1630
05-Aug	*	0000-2400	2200-2400	0000-0530	OFF
06-Aug	*	0000-2400	0000-2400	OFF	OFF
07-Aug		0000-0530	0000-2400	OFF	0800-1630
08-Aug		OFF	0000-2400	2200-2400	0800-1630
09-Aug		OFF	0000-0530	0000-2400	0800-1630
10-Aug		2200-2400	OFF	0000-2400	0800-1630
11-Aug		0000-2400	OFF	0000-2400	0800-1630
12-Aug	*	0000-2400	2200-2400	0000-0530	OFF
13-Aug	*	0000-2400	0000-2400	OFF	OFF
14-Aug		0000-0530	0000-2400	OFF	0800-1630
15-Aug		OFF	0000-2400	2200-2400	0800-1630
16-Aug		OFF	0000-0530	0000-2400	
17-Aug		2200-2400	OFF	0000-2400	
18-Aug		0000-2400	OFF	0000-2400	
19-Aug	*	0000-2400	2200-2400	0000-0530	
20-Aug	*	0000-2400	0000-2400	OFF	
21-Aug		0000-0530	0000-2400	OFF	
22-Aug		OFF	0000-2400	2200-2400	
23-Aug		OFF	0000-0530	0000-2400	
24-Aug		2200-2400	OFF	0000-2400	
25-Aug		0000-2400	OFF	0000-2400	
26-Aug	*	0000-2400	2200-2400	0000-0530	
27-Aug	*	0000-2400	0000-2400	OFF	
28-Aug		0000-0530	0000-2400	OFF	
29-Aug		OFF	0000-2400	2200-2400	
30-Aug		OFF	0000-0530	0000-2400	
31-Aug		2200-2400	OFF	0000-2400	

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Appendix Table A2. Anchor River Weir schedule from July through 15 November, 1989 (continued).

Date	Wknd	Balland	Carver	Kline
01-Sep		0000-2400	OFF	0000-2400
02-Sep	*	0000-2400	2200-2400	0000-0530
03-Sep	*	0000-2400	0000-2400	OFF
04-Sep	*	0000-0530	0000-2400	OFF
05-Sep		OFF	0000-2400	2200-2400
06-Sep		OFF	0000-0530	0000-2400
07-Sep		2200-2400	OFF	0000-2400
08-Sep		0000-2400	OFF	0000-2400
09-Sep	*	0000-2400	2200-2400	0000-0530
10-Sep	*	0000-2400	0000-2400	OFF
11-Sep		0000-0530	0000-2400	OFF
12-Sep		OFF	0000-2400	2200-2400
13-Sep		OFF	0000-0530	0000-2400
14-Sep		2200-2400	OFF	0000-2400
15-Sep		0000-2400	OFF	0000-2400
16-Sep	*	0000-2400	2200-2400	0000-0530
17-Sep	*	0000-2400	0000-2400	OFF
18-Sep		0000-0530	0000-2400	OFF
19-Sep		OFF	0000-2400	2200-2400
20-Sep		OFF	0000-0530	0000-2400
21-Sep		2200-2400	OFF	0000-2400
22-Sep		0000-2400	OFF	0000-2400
23-Sep	*	0000-2400	2200-2400	0000-0530
24-Sep	*	0000-2400	0000-2400	OFF
25-Sep		0000-0530	0000-2400	OFF
26-Sep		OFF	0000-2400	2200-2400
27-Sep		OFF	0000-0530	0000-2400
28-Sep		2200-2400	OFF	0000-2400
29-Sep		0000-2400	OFF	0000-2400
30-Sep	*	0000-2400	2200-2400	0000-0530

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Appendix Table A2. Anchor River Weir schedule from July through 15 November, 1989 (continued).

Date	Wknd	Balland	Carver	Kline	
01-Oct	*	0000-2400	0000-2400	OFF	
02-Oct		0000-0530	0000-2400	OFF	
03-Oct		OFF	0000-2400	2200-2400	
04-Oct		OFF	0000-0530	0000-2400	
05-Oct		2200-2400	OFF	0000-2400	
06-Oct		0000-2400	OFF	0000-2400	
07-Oct	*	0000-2400	2200-2400	0000-0530	
08-Oct	*	0000-2400	0000-2400	OFF	
09-Oct		0000-0530	0000-2400	OFF	
10-Oct		OFF	0000-2400	2200-2400	
11-Oct		OFF	0000-0530	0000-2400	
12-Oct		2200-2400	OFF	0000-2400	
13-Oct		0000-2400	OFF	0000-2400	
14-Oct	*	0000-2400	2200-2400	0000-0530	
15-Oct	*	0000-2400	0000-2400	OFF	
16-Oct		0000-0530	0000-2400	OFF	
17-Oct		OFF	0000-2400	2200-2400	
18-Oct		OFF	0000-0530	0000-2400	
19-Oct		2200-2400	OFF	0000-2400	
20-Oct		0000-2400	OFF	0000-2400	
21-Oct	*	0000-2400	2200-2400	0000-0530	
22-Oct	*	0000-2400	0000-2400	OFF	
23-Oct		0000-0530	0000-2400	OFF	
24-Oct		OFF	0000-2400	2200-2400	
25-Oct		OFF	0000-0530	0000-2400	
26-Oct		2200-2400	OFF	0000-2400	
27-Oct		0000-2400	OFF	0000-2400	
28-Oct	*	0000-2400	2200-2400	0000-0530	END DLST
29-Oct	*	0000-2400	0000-2400	OFF	
30-Oct		0000-0530	0000-2400	OFF	
31-Oct		OFF	0000-2400	2200-2400	

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Appendix Table A2. Anchor River Weir schedule from July through 15 November, 1989 (continued).

Date	Wknd	Balland	Carver	Kline
01-Nov		OFF	0000-0530	0000-2400
02-Nov		2200-2400	OFF	0000-2400
03-Nov		0000-2400	OFF	0000-2400
04-Nov	*	0000-2400	2200-2400	0000-0530
05-Nov	*	0000-2400	0000-2400	OFF
06-Nov		0000-0530	0000-2400	OFF
07-Nov		OFF	0000-2400	2200-2400
08-Nov		OFF	0000-0530	0000-2400
09-Nov		2200-2400	OFF	0000-2400
10-Nov		0000-2400	OFF	0000-2400
11-Nov	*	0000-2400	2200-2400	0000-0530
12-Nov	*	0000-2400	0000-2400	OFF
13-Nov		DISMANTLE WEIR		OFF
14-Nov		DISMANTLE WEIR		
15-Nov		DISMANTLE WEIR, TECH EVALUATIONS		

APPENDIX A3

Anchor River weir fish count form

CONFIDENTIAL

STATE/FEDERAL NATURAL RESOURCE DAMAGE ASSESSMENT
DETAILED STUDY PLAN

CONFIDENTIAL

Project Title: INJURY TO PRINCE WILLIAM SOUND HERRING

Study ID Number: Fish/Shellfish Study Number 11

Lead Agency: State of Alaska, ADF&G;
Commercial Fish Division

DRAFT

Cooperating Agency(ies): Federal: NOAA, OCSEAP/MMS, USFS
State: DNR

Principal Investigator: Evelyn Biggs, Fishery Biologist

Assisting Personnel:

Date Submitted: October 5, 1989

	Signature	Date
Principal Investigator:	Evelyn D. Biggs	9/25/89
Supervisor:	John A Bleasby	9.26.89
OSIAR Senior Biometrician:		
OSIAR Program Manager:	Kelly Holman	10-13-89
OSIAR Director:	for Michael R. Mann	10-17-89

INTRODUCTION

The oil spill in Prince William Sound coincided with the annual migration of Pacific herring to near-shore spawning areas. Approximately 30% of the traditional spawning area in Prince William Sound is located within contaminated areas. Additionally, the majority of adult spawning herring and the newly hatched juveniles will traverse the affected area.

It is hypothesized that the oil spill will adversely impact adult fish through direct mortality, food shortages, slowed growth, and a possible reduction in fecundity. In addition, herring eggs have been shown to be particularly susceptible to hydrocarbon contamination. Possible effects include reduced egg survival, reduced hatching success, and reduced viability of fry. Any of these factors have the capacity to reduce the sustainable yield from herring fisheries in Prince William Sound (worth over 12 million dollars in 1988).

The goal of this project is to determine whether the Exxon-Valdez oil spill will have a measurable impact on populations of Pacific herring *Clupea harengus* in Prince William Sound. Accurate and precise estimates of population abundance, age structure, weight, and length composition data are needed to accomplish this objective. In addition, the direct effects of oil contamination on spawning success and egg survival will be determined.

OBJECTIVES

1. Expand the normal sampling of herring populations in Prince William Sound to increase the precision of herring abundance, age composition, weight, sex ratio, and fecundity estimates. Specifically we intend to:

   Estimate the biomass of the spawning stock of herring in Prince William Sound such that the estimate is within ± 25% of the true value 95% of the time;

   Estimate the age, weight, length (AWL), and sex composition of herring in Prince William Sound during 1989 such that age composition estimates are within ± 5% of their true values 95% of the time;

2. Document the occurrence of herring spawn in oiled and non-oiled areas.

3. Estimate hydrocarbon contamination of, and physiological impacts on, adult herring by analyzing tissue samples for hydrocarbon and histopathological analyses from herring in oiled and non-oiled areas. Specifically, we intend to:

   Test the hypothesis that the level of hydrocarbons in herring tissues is not related to the level of oil contamination of the area from which the herring were sampled. The experiment is designed to detect

a difference of 1.6 standard deviations in hydrocarbon content with the probability of making a type I and type II error of 0.05 and 0.1, respectively.

Estimate the presence and type of damage to tissues and vital organs of herring sampled from oil-impacted and unimpacted areas.

Test the hypothesis that the level of hydrocarbons in herring eggs is not related to the level of oil contamination of the area from which the herring were sampled. The experiment is designed to detect a difference of 1.6 standard deviations in hydrocarbon content with the probability of making a type I and type II error of 0.05 and 0.1, respectively.

4. Estimate the proportion of dead herring eggs in oiled and non-oiled areas.

5. Estimate the hatching success, viable hatch, and occurrence of abnormal larvae by collecting herring eggs from oiled and non-oiled areas and rearing them under laboratory observation.

6. Identify potential alternative methods and strategies for restoration of lost use, populations, or habitat where injury is identified.

METHODS/DATA ANALYSIS

This project will be conducted in three parts: (1) Herring spawn deposition estimation; (2) Herring age, weight, length, growth, and fecundity estimation; and (3) Herring egg survival estimation.

Spawn Deposition Study

The management of the Prince William Sound herring stock is based on a harvest policy established by the Alaska Board of Fisheries which specifies a maximum 20% exploitation rate for the combined harvest of all herring fisheries. The allowable harvest is based on biomass estimates established the previous year modified by the expected growth and survival over the year. While aerial surveys were used to estimate biomass from 1973-87, spawn deposition surveys were performed in 1983 (Jackson and Randall 1983) and 1984 (Jackson and Randall 1984), and began to be used as the primary biomass estimate in 1988 (Biggs and Funk 1988). Aerial surveys are easier to perform than spawn deposition surveys, but aerial survey biomass estimates are not as reliable because of the varying visibility of herring schools from the air and because the residence time of herring schools on the spawning grounds is unknown. Estimates of precision are not available for aerial survey biomass estimates. The Alaska Department of Fish and Game continues to conduct an annual aerial survey of spawning biomass (Commercial Fisheries Division project number TF-300) to provide inseason indicators of run timing and location and to collect information on the timing and distribution of spawning activity that is used for planning the spawn deposition survey.

This project represents an augmented program to assess the Prince William Sound herring stock's response to the oil spill of the Exxon Valdez. The original goal of the 1989 herring spawn deposition survey was to estimate the spawning biomass with a precision such that the biomass estimate would be within ± 28% of the true biomass estimate 95% of the time under optimal survey conditions. Fishery managers determined that this level of precision was acceptable for estimating exploitation rates and forecasting future abundance. If weather or other logistic problems hampered the spawn deposition survey sampling effort, fishery managers were willing to tolerate reduced precision. The oil spill of the Exxon Valdez introduced a potential new and unknown level of mortality on herring stocks. The accuracy and precision of estimates of stock abundance need to be assured from both oiled and unoiled areas (as reflected in objective 1 and 2). The opportunity to estimate herring biomass with spawn deposition surveys is only available during a relatively narrow two week window. After the oil spill, the number of divers involved in the survey was increased to assure that even if weather problems restricted the available sampling time, sufficient numbers of transects could still be performed. The number of transects was also increased to provide a level of precision such that the biomass estimate would be within ± 25% of the true biomass 95% of the time. The amount of time devoted to skiff surveys of spawning areas was also increased. Skiff survey delineation of spawning area boundaries helps to increase the level of precision of spawn deposition surveys and provides important documentation of the occurrence of herring spawn in oiled and unoiled areas.

Study Design:

The aerial survey project (Commercial Fisheries Division project number TF-300) will provide a map indicating the general location of herring spawning areas. A skiff survey will then delineate the boundaries of each spawning area in more detail. Transects will be placed perpendicular to the shoreline at locations selected randomly from the shoreline maps of spawning areas. Divers will swim along the transects and systematically place 0.1 m^2 quadrats at 5 m intervals. Divers will estimate the total number of eggs in each quadrat. All egg-containing vegetation will be removed from a subset of the quadrats for later enumeration of the number of eggs in a laboratory procedure. These enumerated egg counts will be used to correct bias in diver-estimated egg counts and estimate the precision of the diver estimates. The survey design is described in detail by Biggs and Funk (1988), and follows closely the two-stage sampling design of similar surveys in British Columbia (Schwiegert et al. 1985), and in Southeast Alaska (Blankenbeckler and Larson 1982, 1987). The surveys use random sampling at the first stage (transects), and systematic sampling at the second stage (quadrats within transects). Random sampling in the second stage is not feasible because of underwater logistical constraints (Schwiegert et al. 1985). In addition to the two-stage design, the survey is stratified by four areas within Prince William Sound (Northeast, North Shore, Naked Island and Montague), because of the geographic separation of these areas and the potential for herring in these areas to be discrete stocks.

Mean egg densities along each transect will be combined to estimated an overall average egg density. The observed widths of the spawning bed along each of the transects will be used to estimate the average spawning bed width. The average

width, average density, and total spawning bed shoreline length from the skiff survey will be used to estimate the total number of eggs deposited in each of four area strata established within Prince William Sound. Using the average fecundity and sex ratio derived from the AWL sampling portion of this project, the total number of eggs deposited will be converted into population numbers and biomass. Based on the variances obtained during the 1988 survey, 160 transects would be needed to insure that the estimated biomass would have a 95% chance of being within 25% of the true biomass.

Sampling Procedure:

The general locations of spawning activity will be derived from visible milt observed in the water column during scheduled aerial surveys (Commercial Fisheries Division Project Number TF-300). This information will be compiled and summarized on maps showing spawning locations and the number of days on which milt was observed.

Using this information, the skiff survey crew will visit each spawning area and more precisely determine the boundaries of areas actually containing herring eggs. The skiff survey team will begin mapping several days in advance of the dive team, but they will not begin sampling until several days after spawning has ended to ensure that all egg containing areas are recorded. It is anticipated that the skiff survey crew will operate between April 18 and May 4.

The skiff team will be based out of a salmon gill net vessel with shallow draft capabilities, preferably equipped with a jet drive. Sampling will need to be performed very close to shore at low tide, using a small inflatable raft where necessary. Using an underwater viewing box, grappling hooks, rakes, and observations made of the intertidal area, the boundaries of herring spawning areas will be recorded. The total lineal miles of shoreline containing herring spawn will be determined from the skiff survey maps. The skiff survey crew will deliver a completed spawning ground map of each of the four area strata (Northeast, North Shore, Naked Island, and Montague Island) to the spawn deposition diving crew before the diving crew begins to sample each area.

Diving on herring spawn is not recommended for at least 5 days after spawning activity has ceased because of water visibility problems caused by milt and because large numbers sea lions are usually present. Two days of diver training will be conducted between April 19 and May 6, depending on the exact timing of herring spawning, which will include onboard training dives aboard the diving support vessel R/V Sundance. Eight divers, working in two shifts (morning and afternoon), will be needed to complete the project. Based on the 1988 survey and Southeast Alaska surveys, four teams of two divers can complete about 20 transects per day in favorable weather conditions. Assuming that the diving field trip will be 10-14 days, this should allow the survey design goal of 160 transects to be achieved in unfavorable weather. Four divers will be sampling during each shift working in two 2-person teams from two diving skiffs. The four divers not conducting underwater surveys during a shift will serve as dive tenders, refill SCUBA tanks, and maintain equipment.

The shoreline area containing herring spawn on the map received from the skiff

survey team will be divided into the smallest segments resolvable on the scale of the map (0.1 mile or less). A total of 160 of the shoreline segments will be selected at random from all of the spawn-containing shoreline segments. Each transect will be assigned a number and its location drawn on waterproof field maps that can be taken out in the dive skiff. The dive team leader will determine the exact transect location within the randomly selected shoreline segment by identifying a shoreline feature (tree, rock, cliff etc.) located above the high tide line as the dive skiff approaches the shore, but before bottom profiles, bottom vegetation, or herring spawn are visible from the skiff.

A 0.1 m^2 quadrat constructed of PVC pipe will be used for the sampling frame. A depth gauge and compass will be fastened to the quadrat. Data will be recorded on pre-printed single matte mylar forms attached to PVC clipboards, using a large weighted carpenter's pencil attached to the clipboard. Normally the dive team leader will make egg density estimates and record data while the assistant diver sets and follows the compass course, measures distances, and carries and places the quadrat.

Sampling along the transects will occur in the following manner:

1. A compass course perpendicular to the shoreline at the transect location will be set on the compass attached to the sampling quadrat.

2. The first quadrat will be haphazardly placed within the first 5 meters of spawn by tossing the quadrat.

3. The lead diver will estimate and record the number of eggs in the quadrat. The number of eggs is normally recorded in units of thousands. The vegetation type, percent cover, substrate, and depth are also recorded.

4. The assistant diver will measure four complete 1 m hand-spans offshore, along the compass course. Halfway through the fifth hand-span, the assistant diver will gently toss the quadrat ahead approximately one-half meter and allow it to come to rest. The lead diver then makes another estimate at the new quadrat location.

5. This process continues every 5 meters until the apparent end of the spawn is found. Divers will verify the end of the spawn by swimming at least an additional 20 m past the end of the spawn, unless a steep drop-off is encountered.

Appendix A.1 shows the codes for vegetation types that are encountered in Prince William Sound. More than one may be present in the quadrat sampled and the three most common are to be recorded on the data forms. Appendix A.2 shows the data sheets that are to be used in the underwater surveys. Percent cover is a simple estimate of the percentage of plant cover that exists within the quadrat sampled (e.g., if half the area is covered, the cover is 50%).

Approximately every fifth quadrat will be used as a special diver calibration sample. Both divers will estimate the number of eggs in the quadrat in a manner such that neither can see the other's estimate. Divers will attempt to remove

all egg-containing vegetation and scrape eggs off rock substrate, placing the material in numbered mesh bags. A sample size goal of 80 calibration samples per diver was established, including 20 in each of four vegetation categories (eelgrass, fucus, large brown kelp, hair kelp), based on 1988 survey results. Calibration samples should also be spread over a wide range of egg densities. The spawn deposition project leader will track the number of samples collected by each diver by vegetation group and density to ensure that sufficient calibration samples are taken in each category. Upon completing a dive shift, calibration sample material will be removed from the numbered mesh bags and placed in nalgene ziploc bags. Gilson's solution will be poured over the sample so that all material is completely immersed. A label will be made for each sample (preferably in pencil on mylar) containing the transect number, both diver's estimates, date, and vegetation type. Five or 6 calibration sample bags can be stored a 5 gal. plastic bucket. Samples should not be stacked over one another to prevent spilling and mixing. Procedures for the enumeration of the number of eggs in each calibration sample are described in Appendix B.1. The formulas used to prepare Gilson's solution and the other chemicals used for sample processing are also listed in Appendix B.1.

Data Analysis:

Biomass Estimation

The 1989 spawn deposition survey was patterned after the 1988 spawn deposition survey in Prince William Sound (Biggs and Funk 1988). The overall biomass estimator is:

$$B = \frac{(T \cdot B')}{(1 - R)} \quad ; \qquad (1)$$

where:

- B = estimated spawning biomass in tonnes,
- T = estimated total number of eggs (billions) deposited in an area,
- B' = estimated tonnes of spawning biomass required to produce one billion eggs, and
- R = estimated proportion of eggs disappearing from the study area from the time of spawning to the time of the survey.

The estimates for T and B′ are derived from separate sampling programs and are thus independent. Ignoring the unknown variability in R, the estimated variance for the product of the independent random variables T and B′, conditioned on R is:

$$Var(B|R) = \frac{[T^2 Var(B') + B'^2 Var(T) - Var(T) \cdot Var(B')]}{(1-R)^2} \quad ; \qquad (2)$$

where Var(B′) is an unbiased estimate of the variance of B′ and Var(T) is an unbiased estimate of the variance of T (Goodman 1960).

Total Number of Eggs (T)

The total number of eggs deposited in an area was estimated from a two-stage sampling program with random sampling at the primary stage, followed by systematic sampling at the secondary stage, using a sampling design similar to that described by Schwiegert et al. (1985). In computing variances based on the systematic second stage samples it is assumed that eggs are randomly distributed in spawning beds with respect to the 0.1 m^2 sampling unit. While this assumption was not examined, in practice the variance component contributed by the second sampling stage was much smaller than that contributed by the first stage, so that violations of this assumption would have little effect on the overall variance. The total number of eggs (T), in billions, in an area was estimated as:

$$T = N \cdot \hat{y} \cdot 10^{-6} \; ; \qquad (3)$$

where:

L = the shoreline length of the spawn-containing stratum in meters,
N = L/$\sqrt{0.1}$ = the total number of possible transects,
$\sqrt{0.1}$ = 0.3162 m = width of transect strip,
$\hat{y}$ = average estimated total number of eggs (thousands) per transect, and
10^{-6} = conversion from thousands to billions of eggs.

The average total number of eggs per transect strip (in thousands) was estimated as the mean of the total eggs (in thousands) for each transect strip using:

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

where:

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

n = number of transects actually sampled,

i = transect number,

$M_i = w_i/\sqrt{0.1}$ = number of possible quadrats in transect i,

w_i = transect width in meters, and

$\bar{y}_i$ = average quadrat egg count in transect i (in thousands of eggs).

The average quadrat egg count within a transect, $\bar{y}_i$, was computed as:

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

where:

j = quadrat number within transect i,

m_i = number of quadrats actually sampled in transect i, and

y_{ij} = adjusted diver-estimated egg count (in thousands of eggs) from the diver calibration model for quadrat j in transect i.

The variance of T is similar to that given by Cochran (1963) for three stage sampling with primary units of equal size, although in this case the expression is modified because the primary units (transects) do not contain equal numbers of secondary units (quadrats), and the variance term for the third stage comes from the general linear model used in the diver calibration samples:

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

where: $s_1^2 = \frac{\sum_{i=1}^{n} (\hat{y}_i - \hat{y})^2}{n-1}$ = variance among transects,

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

$s_3^2 = \sum_{i=1}^{n} \sum_{j=1}^{m_i} Var(y_{ij})$ = sum of the variances of the individual predicted quadrat egg counts from the diver calibration model,

$f_1 = \frac{n}{N}$ = proportion of possible transects sampled, and

$f_2 = \frac{m_i}{M_i}$ = proportion of quadrats sampled within transects (same for all transects).

Diver Calibration

Diver observations of vegetation species will be aggregated into four vegetation categories based on structural and phylogenetic similarities of plants in the quadrat: eelgrass, fucus, hair kelp, and large brown kelp (Appendix A.1). Diver estimates of egg numbers are approximately proportional to laboratory-enumerated counts, but systematic biases in the diver estimates can be accounted for by vegetation type and density (Biggs and Funk 1988). Individual diver effects were not significant in the 1988 survey, but potential differences among individual divers will be examined. The basic form of models used to account for biases in diver observations is:

$$Y_{ijk} = e^{\alpha} \cdot e^{D_j} \cdot e^{V_k} \cdot X_{ijk}^{\beta_{jk}} \cdot e^{\epsilon} \quad ; \qquad (7)$$

where:

α is a constant,

D_j are parameters representing the effect of j^{th} diver,

V_k are parameters representing the effect of the k^{th} vegetation type,

β_{jk} are parameters controlling the functional form of the relationship between the diver estimate and laboratory-enumerated egg count for diver j in vegetation type k,

Y_{ijk} is the i^{th} laboratory egg count in the vegetation-diver stratum jk,

X_{ijk} is the i^{th} diver estimate in vegetation-diver stratum jk, and

ϵ is a normally distributed random variable with mean 0 and variance σ^2.

A multiplicative-effect model was chosen because relative estimation errors were expected to change with egg density. The distribution of laboratory-enumerated egg counts for a given diver estimate was positively skewed in the 1988 survey (Biggs and Funk 1988), so that the logarithmic transformation used to estimate the parameters of the multiplicative-effect model also stabilized the variance and corrected the skewness of the egg density estimates. After a logarithmic transformation model 7 becomes:

$$\log_e(Y_{ijk}) = \alpha + D_j + V_k + \beta_{jk} \cdot \log_e(X_{ijk}) + \epsilon \qquad (8)$$

The parameter β_{jk} is the slope of the relationship between the logarithm of the diver estimate and the logarithm of the laboratory-enumerated egg count. In logarithmic form, the model comprises a linear analysis of covariance problem with two factor effects (vegetation and diver) and 1 covariate (diver-estimated egg number). The SAS procedure for general linear models (SAS 1987) will be used to obtain least squares estimates of parameters and evaluate variance components. In addition to the two factor effects and one covariate, terms for

diver-vegetation group interactions, density-vegetation group interactions and density-diver interactions will be considered in the analysis of covariance. Three-way and higher level interaction effects will not be considered because the objective is to derive a simple model with a relatively small number of parameters. Backward stepwise procedures will be used to determine subsets of the six effects that explain the maximum amount of variability in the data with the smallest number of parameters. During the backward stepwise procedures, effects will be included or eliminated from the model based on the probability level of F ratios for partial sums of squares.

Translation of the predicted values from the logarithmic model, equation (8) back to the original scale, equation (7) requires a correction for bias. The bias in the expected value of Y_{ijk} is $\exp(\frac{1}{2}\sigma^2)$ when the true variance of Y_{ijk}, σ^2, is known, and Laurent (1963) gives an exact expression for the bias correction that incorporates additional terms when σ^2 is estimated from a sample. For the diver calibration data, the biases in estimating σ^2 from a sample were less than 0.05% (Biggs and Funk 1988) so expected values for Y_{ijk} were estimated from:

$$E(Y_{ijk}) = e^{\alpha} \cdot e^{D_j} \cdot e^{V_k} \cdot e^{B_{jk}} \cdot X_{ijk} \cdot e^{\frac{1}{2}s^2} ; \qquad (9)$$

where s^2 was the mean squared error from the general linear model. The variance of individual predicted Y_{ijk} was estimated from:

$$Var(Y_{ijk}) = [e^{(2Y_{ijk} + \sigma^2)}] \cdot [e^{\sigma^2} - 1] . \qquad (10)$$

This expression is appropriate when σ^2 is known (Laurent 1963), although it should be possible to use s^2 instead of σ^2 without correction for bias, because the bias introduced into estimates of the mean when s^2 was used for σ^2 were found to be small by Biggs and Funk (1988).

Spawning Biomass per Billion Eggs (B')

Catch sampling programs will be used to estimate the relationship between spawning biomass and egg deposition. The tonnes of spawning biomass required to produce one billion eggs (B') will be estimated as:

$$B' = \frac{\bar{W} \cdot S}{F(\bar{W}_f)} \cdot 10^3 ; \qquad (11)$$

where:

$\bar{W}$ = estimated average weight in grams of all herring (male and female) in

the spawning population in an area,

S = estimated ratio of total spawning biomass (male and female) to female spawning biomass,

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

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

Because average weight, sex ratio and fecundity are all estimated from essentially the same sampling program, the estimates are not independent. The variance of B' is approximately:

$$\begin{aligned} Var(B') = (10^3)^2 \{ & [S/F(\bar{W}_f)]^2 \, Var(\bar{W}) \\ & + [\bar{W}/F(\bar{W}_f)]^2 \, Var(S) \\ & + [\bar{W}S/F(\bar{W}_f)^2]^2 \, Var(F(\bar{W}_f f) \\ & + 2Cov(\bar{W},S) \, [S/F(\bar{W}_f)] \, [\bar{W}/F(\bar{W}_f)] \\ & - 2Cov[\bar{W},F(\bar{W}_f)] \, [S/F(\bar{W}_f)] \, [\bar{W}S/F(F(\bar{W}_f)^2] \\ & - 2Cov[S,F(\bar{W}_f)] \, [\bar{W}/F(\bar{W}_f)] \, [\bar{W}S/F(\bar{W}_f)^2] \} \end{aligned} \quad (12)$$

Because S was estimated from pooled AWL samples and in two of the areas S was estimated from a single AWL sample, it was not possible to estimate the covariance terms containing S, ($Cov(\bar{W},S)$ and $Cov[S,F(\bar{W}_f)]$), so these terms were not included in the estimate of $Var(B')$. These covariance terms probably contribute a negligible amount to $Var(B')$, because the term involving $Cov[\bar{W},F(\bar{W}_f)]$ was very small.

Mean Weight and Sex Ratio

Mean weight and sex ratio will be estimated from age-weight-length (AWL) samples collected from a purse seine fishing vessel charted by ADF&G. In a normal year, AWL samples would have been obtained both from the commercial catch and ADF&G test fishing conducted before or after commercial openings by ADF&G and by commercial vessels. For 1989, the chartered purse seine vessel will be directed to obtain AWL samples representative of the spawning population in each of the spawn deposition summary areas (Valdez Arm, North Shore, Naked Island, and Montague Island). The approximate timing of peak herring spawning in each summary area will be determined from aerial survey sightings of milt and herring schools. All fish from AWL samples taken during the time of peak spawning in each area will be pooled to obtain estimates of mean weight and sex ratio for each summary area. Average weights and sex ratios for all of Prince William Sound will be estimated as a weighted average of the estimates from each of the

areas, weighting by the spawn deposition biomass estimate in each area.

The sex ratio estimate, S, is computed as the ratio of the number of fish of both sexes in the AWL samples to the number of females. The binomial distribution is applicable to estimating the proportion, p, of females in AWL samples, where S = 1/p. The variance of S is then given by:

$$Var(S) = \frac{S'(S-1)}{n}, \qquad (13)$$

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

Egg Loss

Before the extensive use of diving surveys of herring egg deposition, estimates of herring egg loss between the time of spawning and the time of egg deposition were relatively high. Montgomery (1958) estimated that in Southeast Alaska egg loss was 25 to 40%, and similar high egg losses were used in the early studies of egg deposition in Southeast Alaska (Blankenbeckler and Larson 1987). However, Haegele et al. (1981) argued that these estimates were high because most spawn was thought to be intertidal before the advent of diving surveys and that intertidal predation and wave loss is probably higher than subtidal. Haegele et al. (1981) estimate egg loss to be approximately 10%, primarily because of predation and wave action loosening the eggs from the substrate during storms. Since the timing of diver surveys following spawning is similar in British Columbia, Southeastern Alaska and Prince William Sound, the 10% egg loss used in British Columbia and Southeast Alaska (W. Blankenbeckler, Alaska Department of Fish and Game, Ketchikan, personal communication) was used for the 1988 Prince William Sound egg deposition survey.

Herring Age, Weight, Length, Growth, and Fecundity Estimation Study

Because of the oil spill all commercial fisheries for herring in Prince William sound were closed in 1989. The normal management of these fisheries would have included a state vessel located near the fishery that would function as headquarters for management and research staff to conduct test fishing. Normal test fishing would have involved placing Department staff on board fishing vessels to sample their catch for age, weight, length, (AWL), fecundity, and roe maturity information. This information is necessary to estimate spawning biomass and spawn deposition, predict future returns for herring, and evaluate the effects of the spill on survival. Fecundity, average weight of females, and sex ratio are also components of the spawn deposition biomass estimator. Because tighter than normal precision for the spawn deposition biomass estimator was needed to evaluate oil spill impacts, more intensive AWL sampling is required. This study was developed in order to provide data that is normally supplied by the commercial fishing vessels; since no fishery occurred, a vessel had to be chartered with a technician to fill in the gap. In addition, sampling intensity will be increased where required to increase precision.

Study Design:

A test fishing purse seine vessel will be contracted from April 4 to 18 to capture fish for AWL sampling. Sampling will begin as soon as concentrations of herring appear in near shore areas that are accessible to purse seines. Spawning migration samples will be taken from most large herring concentrations throughout the Prince William Sound and at periodic intervals by region throughout the major spawning period. The objective is to document the age-sex-size composition of all major biomass concentrations in the general areas of Valdez Arm and the Eastern District, the North Shore, Naked Island, and Montague Island. Results of the aerial survey program (TF-300) will be used to direct test fishing to major biomass concentrations.

Sampling Procedures:

Six to eight samples of herring will be collected using a chartered purse seine vessel each week during the spawning migration (early-April through early May). Each sample will consist of 500 fish, approximately the size recommended by Thompson (1987) to achieve confidence intervals for proportions at age such that 95% of the time the estimated proportions will be within 5% of the true proportions. Age composition samples will be flown to Cordova from the sampling vessel daily for processing. Sampling, processing, and archiving methods are described in detail in the Prince William Sound Herring Catch Sampling Operational Plan, CF-302 (Appendix D.1) and the Prince William Sound AWL Sampling Manual (Appendix D.2). Augmentation of the normal AWL sampling program was needed to collect hydrocarbon, organoleptic, and fecundity samples and to cover the cost of the boat charter to collect herring samples. All age, weight and length data was collected with resources and personnel from the regular AWL program (non-oil spill related).

The following data will be collected for each herring sampled:

1. Sex determined by examination of gonads;
2. standard length (in mm);
3. weight (in grams); and;
4. scale collected to determine age;
5. date of capture, fishing district and subdistrict, local name for the location fished, name of the fishing vessel and gear type; and,
6. data form number and the fish number for that form.

In addition, a subsample of fish will be collected to estimate fecundity. The average fecundity at the average female weight ($F(\bar{W}_f)$ from expression 11) is one component of the spawn deposition survey biomass estimator. The spawn deposition survey attempts to estimate spawning biomass with 95% confidence intervals of no more than ± 25% of the biomass point estimate. If fecundity sampling is to contribute no more than 1% to the confidence interval width, a sample of 85 females of exactly the average weight of females in spawning population is needed. Because the average female weight is unknown at the time of sampling, more fish must be sampled over a range of sizes. Based on the

precision of 1988 fecundity sampling, a sample size of 130 fish would be needed to provide the desired level of precision (Fritz Funk, Alaska Department of Fish and Game, personal communication). An additional 100 samples clustered around the average size of females in 1988 will be taken to compare with the past year's data. The average length of a female in the fecundity sample in 1988 was 200 mm. The predicted average weight for the population in 1989 is 127 grams that translates to an average predicted length of 200 to 215 mm. Clustering of samples around the 200 to 220 mm length classes is desirable.

It is also of interest to investigate whether fecundity varies by region because of oil impact. A fecundity sample (400 females) is requested from oil impacted and non-impacted areas. Minimally 20 females from each of the 5 length categories will be collected for a sample size of 100 per stratum. In addition, the size stratum recommended for 1989 to allow comparisons of year and oil effects are:

20 females @ 181-190 mm
60 females @ 191-200 mm
65 females @ 201-210 mm
65 females @ 211-220 mm
20 females @ 221-230 mm.

Area strata are defined as the (1) Valdez Arm and Eastern District, (2) North Shore, (3) Naked Island, and (4) Montague Island Area.

Herring fecundity samples will be collected concurrently with AWL samples (project CF-302). At least five of the individual test purse samples brought in will be subsampled in the following manner. For each 10 mm length class, females will be randomly selected (until the stratum goals are reached) and their roe sacs removed; the AWL number for each female will be recorded and written on individual ziploc bags in which the roe from that fish will be placed (e.g., 20 for 151-160 mm standard length fish, 20 for 161-170 mm). These individually packaged roe samples will then be placed in a larger plastic bag with the sample date and location for cross referencing later while processing data. The lab procedures for processing individual fecundities are listed in Appendix B.2.

The following samples for histopathology and hydrocarbon will also be obtained from each of the four locations (Naked Island, Galena Bay, Cedar Bay, and Stockdale Harbor):

1. Three gut samples for hydrocarbons;
2. Three viscera samples for hydrocarbons;
3. Three muscle samples for hydrocarbons;
4. Three gonad samples for hydrocarbons;
5. Twenty histopathology samples; and
6. Observations as to anignis prevalence (nematode), liver and gall bladder condition, and fullness of gut will be collected for each fish sampled.

Sample sizes and collection strata for hydrocarbon and histopathology are discussed in detail in Appendix E.

Data Analysis:

A linear regression between fecundity and weight provided a reasonable description of fecundity data collected in 1988 (Biggs and Funk 1988). Average fecundity for each area will be estimated from the fecundity-weight relationship using the average female weight in the AWL samples from each area and applied to the spawn deposition biomass estimator ($F(\bar{W}_t)$ in expression 11). The variance of estimated average fecundities is approximated by the variance of predicted means from the fecundity-weight linear regression (Draper and Smith 1981):

$$Var[F(\bar{W}_t)] = s' \left[\frac{1}{n} + \frac{1}{q} + \frac{(\bar{W}_t - \overline{WF})'}{\Sigma(W_i - \overline{WF})'} \right] \qquad (14)$$

where s' is the residual mean square from the fecundity-weight linear regression, $\bar{W}_t$ is the average weight of female fish in the spawning population, $\overline{WF}$ is the average weight of females in the fecundity sample, W_i are the weights of individual females in the fecundity sample, n is the total number of females in the fecundity sample, and q is the total number of females in the AWL sample.

General Linear Model (GLM) extensions of linear analysis of variance (ANOVA) techniques will be used to test for year and area effects in growth and fecundity. Techniques employed in processing herring AWL data are discussed in more detail in the Prince William Sound Herring Caatch Sampling Operation Plan, CF-302 (Appendix D.1).

Egg Survival Study

The oil spill has the potential to cause mortality to herring eggs because herring spawn in intertidal and shallow subtidal areas where their adhesive eggs remain attached to vegetation and other substrates for a three week incubation period. This study was developed to estimate immediate, observable mortality of herring eggs in areas of high, medium, low, and no oil impact.

Study Design:

Five study transects will be established in each of the areas designated as high, medium, low, and no oil impact. The ratio of live to dead eggs will be determined along each transect. Dead herring eggs turn an opaque white color, which is easily identified under low power magnification. Herring spawning locations will be mapped by ADF&G aerial surveys and skiff surveys associated with the spawn deposition project. Shoreline survey data mapped by the Alaska Department of Environmental Conservation (DEC) should be available to determine which of the spawning areas would be suitable locations for high, medium, and low oil impact study areas. Mussel tissue samples will also be collected for hydrocarbon analysis. Mussel tissue hydrocarbon levels may provide a basis for more precise post-stratification of oil impact levels if DEC shoreline maps are unavailable or unsuitable.

A separate project will be conducted by a contractor to the Outer Continental Shelf Environmental Assessment Program (OCSEAP) of the National Oceanic and Atmospheric Administration to examine the survival of herring eggs and larvae

collected in oiled and unoiled areas under laboratory-controlled conditions. Eggs for the laboratory study will be collected from transect locations established by the field egg survival study.

Sampling Procedures:

The egg survival project will be conducted from a vessel capable of housing 4 biologist/technicians in addition to the vessel crew. Sampling will be performed from a 14-18 ft skiff (inflatable boat equivalent to a Mark II Zodiac, 17' Boston Whaler, or 16+ foot aluminum skiff).

Five transects containing herring eggs in each oil contamination category (high, medium, low, none) will be established. The ratio of live to dead herring eggs will be determined along the transects at set depth intervals. Hydrocarbon sampling of mussels and herring eggs will be conducted at the lowest tidal level in which mussels occur. The transects will be resampled every 4 days until hatching (about May 15).

Transects will be established for the duration of the project. Egg-containing shoreline on Naked Island classified as low, medium, and high oil impact from DEC shoreline survey maps and ADF&G aerial herring milt survey maps will be identified. Unimpacted shoreline on the north shore (Granite Pt. to Unakwik Pt.) will be surveyed for potential use as control (unimpacted by oil) sampling sites. Shoreline in each treatment level (high, medium, low, control) will be surveyed by personnel using a skiff to identify suitable study sites. Criteria for selection as a transect will be: (1) herring eggs present, (2) at least a moderate band of mussels present, (3) only one or two herring egg layers present (4) a relatively wide band of herring eggs present (to at least 15', (5) consistency as to exposure level and kelp community with sites in other treatment categories. The presence, number of layers, and depth distribution of herring eggs at potential sites will be determined by sampling with grappling hooks and reconnaissance diving. It is desirable to keep herring egg layers less than three because egg survival is known to be reduced in thickly layered eggs (Hourston et al. 1984).

Sampling locations will be selected by the dive team leader from these areas of potentially suitable shoreline, after initial reconnaissance is completed. The location of each transect will be marked with a spray painted fence post placed above the high tide line. Transects will be oriented perpendicular to the shore following a compass course. During the first dive, sample stations at the +1, 0, -15, and -30 foot depths will be marked underwater with weighted floats anchored by a spike. Station depths will be located using diver's depth gauges, corrected for stage of tide.

Each transect will be sampled every four days. This will result is 4-5 dives along each transect over the course of egg development. Divers will establish the location of mean lower low water (MLLW) at the time of the start of the dive. Divers will attempt to sample 5 transects each day. This should allow the single team of divers to use no more than two tanks of air per day to minimize potential decompression problems.

For the sampling at each station, divers will place a leadline perpendicular

and to the right of the transect sampling location (i.e., parallel to the beach but at +1, 0, -1, -15, or -30 ft depths). The leadline is marked in three places in each of three different colors (red, blue, green) for a total of 9 placements. The length of the leadline and spacing of the ribbons will depend on the substrate patch size but will be no more than 20 m for practical purposes. Divers will select the plant whose stem is closest to the 3 red marks. If no egg containing plants are close to the red marks, blue, then green marks will be used. This step avoids sampler bias in selecting egg samples. Egg-containing vegetation may not be available at the shallow and deep extreme depths. Divers will collect 3 samples of vegetation containing at least 100 eggs at each depth along the transect and place the samples into pre-labelled mesh bags. Samples will be kept moist until eggs can be counted. Numbers of live and dead eggs will be counted within 4 hours of sampling. Counts will be made with a binocular microscope until 100 eggs have been counted from each sample.

The following data will be recorded for each transect:

1. Transect number;
2. Location, description of exposure, plant community;
3. Number of depth strata with herring eggs; and,
4. Treatment category (high, medium, low, no oil-impact).

For each sampling replication of the transect the following data will be recorded:

1. Transect number, date, time in/out, location, treatment level;
2. Air and water temperature, maximum depth; and,
3. Number of live, dead, and other eggs per (bag) vegetation sampled.

Herring eggs and mussels will be collected for hydrocarbon analysis. The protocol for collection, labeling, chain of custody forms, and sample design is discussed in Appendix E. Hydrocarbon samples will be collected at each transect location on the first sampling day. Three samples each of eggs and mussels (6 per transect) will be collected from each sampling location at the lowest location at which mussels occur (approximately 5 feet below MLLW). In addition, one transect each will be selected from the high oil impact and control category for depth stratified sampling. Three samples of mussels will be collected from the 0, -5, and -15 feet level for the two transects. Chain of custody forms will be filled out for each hydrocarbon sample collected.

During one of the repetitions at each transect, egg-containing vegetation will be collected for the laboratory incubation project. The collection of live eggs for the laboratory studies of herring egg hatching success and larval viability is discussed in Appendix F. This project is being performed by Envirocon Pacific Limited of Canada, under contract to OCSEAP. Three samples of vegetation containing at least 300 eggs will be collected at MLLW, -5 ft, and -15 and placed in separate, labelled mesh bags. A technician from the laboratory incubation project will supervise the handling and packing of the eggs for transportation. Eggs must be kept cool and moist. Chartered aircraft will pick up the packaged egg samples each day for transportation to the laboratory. Collection of eggs for the laboratory project from each of the 20 transacts should take 4 days. A total of 180 vegetation samples will be collected (20 transacts, 3 depths, and 3 replicate samples at each depth). The technician

from the laboratory incubation project will supervise the transportation and shipping of the eggs to the laboratory site.

Data Analysis:

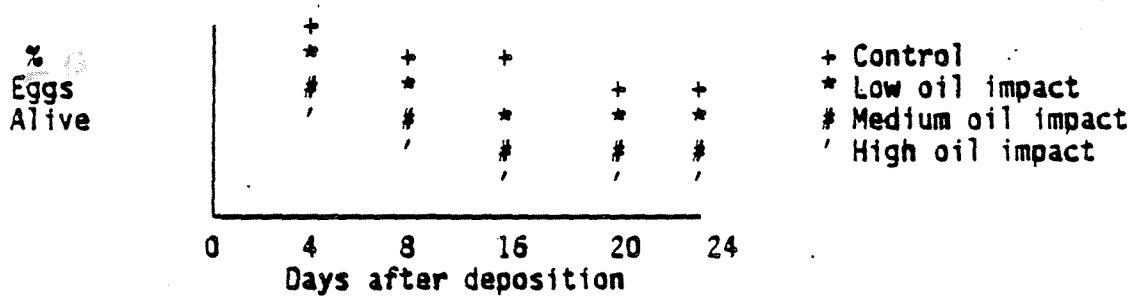
ANOVA and/or general linear model extensions will be used for analyzing differences in egg mortality among sites. A full ANOVA model incorporating all possible factors and interaction effects from depth, treatment (oil impact level), and date would be:

$$Y_{ijkl} = U + A_i + B_j + C_k + (AB)_{ij} + (AC)_{ik} + (BC)_{jk} + (ABC)_{ijk} + \epsilon_{ijkl} \quad (15)$$

Where Y_{ijkl} is the arc sin transformed proportion of live eggs, U is the grand mean, A is the treatment effect, B is the depth effect, and C is the effect for date of observation, and AB, BC, AC, and ABC are interaction effects. The errors, ϵ_{ijkl}, are assumed normally distributed after the arcsin transformation. A general linear model of the form:

$$Y_{ijk} = U + A_i + B_j + C(t) + AB_{ij} + \epsilon_{ijk} \quad (16)$$

will also be investigated where C(t) is a function of two or more parameters of time since egg deposition. Parameters of C(t) may have to be specific to depths or treatments. The general linear model describes the decrease in the ratio of live to dead eggs over time, using time as a covariate, reducing the number of parameters to be estimated in the model. The replicated proportions of eggs and larvae surviving will plotted against time for oil-impacted and control experiments, for example:



SCHEDULES AND REPORTS

Date(s)	Activity
Spawn Deposition Program	
Apr. 18-May 4, 1989	Skiff surveys of spawning areas.
Apr. 19-May 6, 1989	Diver training and transect sampling for spawn deposition.
May-August 1989	Laboratory analysis of samples
September-October 1989	Data Entry and Analysis
November 1989	Report Preparation.
December 15, 1989	Technical reports completed: Pacific herring spawning deposition surveys for Prince William Sound, 1989.
January 15, 1989	Sampling manual and project study plan for 1990 completed.
AWL Sampling Program	
April 4-25	Test fishing and AWL/fecundity sampling
May 1989-January 1990	Laboratory analysis of samples
August-October 1989	Data Entry and Analysis
November-December 1989	Report Preparation.
March 15, 1990	Technical reports completed.
March 30, 1990	Sampling manual and project study plan for 1990 completed.

Egg Survival Study

April 19-May 6 1989	Transect sampling for egg mortality and hydrocarbon analysis
April 20-June 15, 1989	Laboratory studies of oil impacts of herring eggs.
May-August 1989	Laboratory analysis of samples
August-October 1989	Data Entry and Analysis
November-December 1989	Report Preparation.
January 31, 1990	Technical reports completed.
March 30, 1990	Sampling manual and project study plan for 1990 completed.

PROJECT BUDGET

PROJECT BUDGET [1]

Line Item	Category	Budget
100	Personnel Services	$ 85,000
200	Travel	$ 9,500
300	Contractual	$148,000
400	Commodities	$ 19,000
500	Equipment	$113,000
700	Grants	$ 0
Total		$374,500

[1] Budget is for all activities performed from March 27, 1989 to February 28, 1990.

FUNDED PERSONNEL [1]

Class	PCN	Name	PFT_mm	SFT_mm
FB II	11-1346	Biggs	9.0	
FT III	11-1394	Norman		3.5
FT II	11-1532	Bracken		2.5
FT I	11-1925	Carpenter		2.0
BIOM II	11-	Baker	1.0	
FT III	11-NP*	McNeil		1.5
FT II	11-NP*	Walters		2.0
BIOM II	11-1928	Geiger	1.0	

[1] Additional permanent staff funded under existing Commercial Fisheries Projects will assist with sampling and data analysis. They include: Tim Baker Biom. I (3.5 mm); Evelyn Biggs FB II (5 mm in 1989); Fritz Funk, Biom . II/Alternate Diver (3 weeks); Tim Minicucci FB II/Calibrated Diver (2 weeks); Doug Jones FB II/Alternate Diver (2 weeks); Tom Rutecki, NMFS/Alternate Diver (2 weeks); John Hamilton NMFS/Alternate Diver (2 weeks); Dennis Blankenbeckler FB III/Calibrated Diver (2 weeks); Wade Loofbourrow Skipper (3 weeks); and Vessel 1st mate (3 weeks).

* NP is non-permanent employees.

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APPENDICES

Appendix A.1. Vegetation and substrate codes used in the 1988 Prince William Sound egg deposition survey, and vegetation summary categories.

Vegetation Codes	Veg./Substrate Name	Vegetation Group
ag	Agarum	LBK
ala	Alaria	LBK
alg	algae	LBK
bf	brown filamentous	HRK
bhk	brown hair kelp	HRK
bol	boulders	FUC
cob	cobble	COB
cor	corraline	LBK
cos	Costaria	LBK
ee	elephant ear	LBK
eel	eelgrass	EEL
eg	eelgrass	EEL
fg	Filamentous green	HRK
fir	fir weed	LBK
frn	fern (Odonthalia)	HRK
fu	Fucus	FUC
gf	green filamentous	HRK
gl	green leaf	HRK
gr	gravel	FUC
hr	hair kelp	HRK
hrk	Hair kelp	HRK
lam	Laminaria sp.	LBK
lbk	large brown kelp	LBK
le	loose eggs	EEL
mix	mixed	MIX
mud	mud	EEL
ral	red algae	LBK
rb	ribbon kelp	LBK
rc	red cup (Constantinea)	LBK
rck	rock	FUC
rf	red filamentous	HRK
rh	red hair kelp	HRK
rhk	red hair kelp	HRK
rho	Rhodymenia	HRK
rib	ribbon kelp	LBK
rk	rock	FUC
rl	red leaf	LBK
rrb	red ribbon kelp	LBK
sd	sand	EEL
ul	Ulva	FUC
ulv	Ulva	FUC
yz	Laminaria yezoensis	LBK

Appendix B.1. Laboratory methods for calibration samples and chemical recipes.

Calibration samples collected in the manner described in the text will be processed with the following methods:

1.) Decant the Gilson's solution from the sample bags.

2.) Add KOH to the sample bag and thoroughly mix through the sample. Allow the sample to soak for 1.5 hours in a hot water bath, which accelerates the digestive hydrolysis that will take place. Eelgrass and fucus samples may take longer than hair kelps and LBK (large brown kelps). Monitoring the digestive process by gently manipulating the sample bags will give insight to the timing of the soak required.

3.) Drain off the KOH, by decanting, estimating the egg loss, and place the sample in a one gallon plastic bucket.

4.) Repeated washing of the sample with cold water will loosen many of the attached eggs. The sample can be washed by pouring it, eggs and all, into an appropriately sized sieve, pouring cold water over the top and allowing the dissolved kelp samples to rinse through the mesh.

5.) Any eggs left attached must be removed from the substrate by careful manual scraping. The loose eggs must be clean from debris for accurate volumetric analysis. Any eggs lost during the cleaning process must be estimated and recorded.

6.) The loose, clean eggs are then poured back in the labeled, rinsed sample bags and 1.0 Normal bufferred formal saline solution is pouring into the bags covering the eggs generously. The eggs are allowed to soak in the saline for 24 hours to insure standardized volumetric displacement.

7.) The preliminary step in quantitative analysis is to determine the standard displacement of 1,000 eggs. This is done by hand counting 1,000 eggs from the sample and measuring the volume in an appropriately sized graduated cylinder (10 ml size works well).

8.) The whole sample is them measured volumetrically and the total number of eggs in the sample estimated by simple proportion. This figure is expanded by 10 to determine the density in eggs/meter squared since the sample is taken from a 1/10th of a meter squared.

9.) The lab determined actual counts are then compared to the diver estimates for that sample and a diver calibration correction factor can then be calculated.

10.) Generally, two people can work up 6-8 samples per day.

Gilson's Solution:

For five gallons (in 5 gallon plastic fuel containers), mix

380 ml Formalin
340 ml Glacial Acetic Acid
285 ml 70/70 Nitric Acid
1140 ml Anhydrous Alcohol or 1890 ml 60% Alcohol(EtOH)
Pour water in to make five gallons.

Note: adding a little water to the container prior to mixing reduces fumes produced. Mixing should be done under a hood if possible and if not, near an exhaust fan.

1 Normal KOH (Potassium Hydroxide)

For 1 gallon, mix 224 g. KOH crystals to 4000 ml of water.
For 1 liter, 56.11 g. KOH are mixed with 1 liter of water.

1 Normal Bufferred Formal Saline:

Add 58.44 g. NaCl (pickling salt, table salt has additives) per liter of water or 233.76 g. for 4 liters of water. The saline solution is bufferred by adding the appropriate amounts of sodium bicarbonate (baking soda) per liter of solution:

add2.5 g. for 6.5+ pH
2.0 g. for 6.5 pH
1.5 g. for 6.0-6.5 pH
1.0 g. for -6.0 pH

Add 1 part 30% formalin to 10 parts buffered saline to prevent egg decomposition.

Appendix B.2. Laboratory methods for fecundity samples.

The lab procedures are for mature, fully developed herring ovaries that contain very little membrane and contain fully developed roe.

1.) Remove from the freezer only the samples that can be processed in a single day; samples can be removed the night before if they will be processed the following morning. Generally 15-20 samples can be processed per day per person.

2.) The roe is removed from its individual zip-loc bag and weighed on a scale to the nearest .01 gram. All ice and debris should have been removed. The sample location, date and AWL number should be recorded for each sample.

3.) Four subsamples are then removed from random locations from each roe sample that each weight from .10 to .30 grams (this will be around 200 eggs plus) and placed in labeled petri dishes. The weight from each subsample is carefully recorded. Gilson's solution is then poured into the petri dishes and allowed to soak for 5 minutes or more to loosen the eggs from the connective membrane.

4.) Once the eggs are loose, the Gilson's can be decanted off and the eggs from each subsample can be counted with a tally whacker and recorded.

5.) The mean and standard deviation of the egg count per weight is then calculated for the four subsamples and the total number of eggs in the roe sample is back calculated by simple proportion. The standard deviation of the egg total or fecundity can be calculated by multiplying the total sample weight by the standard deviation of the mean of the four subsamples egg count per unit weight (Brannian, 1988).

6.) The weights and egg counts will be recorded in a data notebook and the data will be entered onto a Lotus spreadsheet where the final calculation of fecundity and standard deviation can be calculated.

Appendix A.1. Vegetation and substrate codes used in the 1988 Prince William Sound egg deposition survey, and vegetation summary categories.

Vegetation Codes	Veg./Substrate Name	Vegetation Group
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4.) Repeated washing of the sample with cold water will loosen many of the attached eggs. The sample can be washed by pouring it, eggs and all, into an appropriately sized sieve, pouring cold water over the top and allowing the dissolved kelp samples to rinse through the mesh.

5.) Any eggs left attached must be removed from the substrate by careful manual scraping. The loose eggs must be clean from debris for accurate volumetric analysis. Any eggs lost during the cleaning process must be estimated and recorded.

6.) The loose, clean eggs are then poured back in the labeled, rinsed sample bags and 1.0 Normal bufferred formal saline solution is pouring into the bags covering the eggs generously. The eggs are allowed to soak in the saline for 24 hours to insure standardized volumetric displacement.

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8.) The whole sample is them measured volumetrically and the total number of eggs in the sample estimated by simple proportion. This figure is expanded by 10 to determine the density in eggs/meter squared since the sample is taken from a 1/10th of a meter squared.

9.) The lab determined actual counts are then compared to the diver estimates for that sample and a diver calibration correction factor can then be calculated.

10.) Generally, two people can work up 6-8 samples per day.

Gilson's Solution:

For five gallons (in 5 gallon plastic fuel containers), mix

380 ml Formalin
340 ml Glacial Acetic Acid
285 ml 70/70 Nitric Acid
1140 ml Anhydrous Alcohol or 1890 ml 60% Alcohol(EtOH)
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Note: adding a little water to the container prior to mixing reduces fumes produced. Mixing should be done under a hood if possible and if not, near an exhaust fan.

1 Normal KOH (Potassium Hydroxide)

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2.0 g. for 6.5 pH
1.5 g. for 6.0-6.5 pH
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4.) Once the eggs are loose, the Gilson's can be decanted off and the eggs from each subsample can be counted with a tally whacker and recorded.

5.) The mean and standard deviation of the egg count per weight is then calculated for the four subsamples and the total number of eggs in the roe sample is back calculated by simple proportion. The standard deviation of the egg total or fecundity can be calculated by multiplying the total sample weight by the standard deviation of the mean of the four subsamples egg count per unit weight (Brannian, 1988).

6.) The weights and egg counts will be recorded in a data notebook and the data will be entered onto a Lotus spreadsheet where the final calculation of fecundity and standard deviation can be calculated.

Appendix C. Pre-season diver check-off list for herring spawn deposition Surveys.

All divers with no exception must be state certified in order to participate in the underwater surveys. The following conditions must be met prior to conducting surveys:

1.) All divers must be certified through a nationally recognized training organization such as P.A.D.I., N.A.U.I., YMCA, S.S.I., or N.A.S.D.S.

2.) All divers must undergo an annual physical prior to the season, which includes parameters outlined in the State SOP manual, Appendix E, Table 1.

3.) All divers must complete a pre-season check-out dive, using department or personal equipment with one of the team leaders and the RDO (Regional Dive Officer).

4.) All divers must have a Letter of Authorization to dive from the State DSO (Dive Safety Officer) and signed by the commissioner. The format for this letter is outlined in Appendix B. of the State SOP. The letter will list the diver as a Diver Trainee, Limited, or Advanced, the qualifications for which are outlined in the SOP manual.

5.) All divers must maintain a log that lists departmental and non-departmental dives made, including location, date, maximum depth, bottom time, water visibility, purpose of dive and any other information of interest. These logs will be kept on file by the RDO.

6.) Survey divers will report any equipment that is in need of maintenance or repair immediately to the RDO so that maintenance can be performed and recorded.

7.) All divers will read and be familiar with the Department Dive SOP Manual as well as the Emergency Dive Accident Procedure Manual provided with the seasons operational manual notebook.

8.) All divers will understand that non-compliance with Department SOP, including the waivers allowed in the annual regional dive memo, and disregard of safety procedures while conducting dive operations will be considered grounds for immediately suspension or dismissal.

APPENDIX D.1

Prince William Sound Herring Catch Sampling Operational Plan (Project CF-302)

FN: PWSHERCT.WP5

9.15.88 PWR Wordperfect Version S.Sharr Draft 1/24/88 4:30

ALASKA DEPARTMENT OF FISH AND GAME
DIVISION OF COMMERCIAL FISHERIES

PROJECT OPERATIONAL PLAN

Title: Prince William Sound Herring Catch Sampling

Project Leader: Sam Sharr

PCN: 1210

Date Submitted:

Region: II

Fishery Unit: Prince William Sound Herring

Yellow Book Project No.: CF-302

File Name: PWSHERCT.WP

Fiscal Year: 1989 **Total Project Cost:** $10,100

APPROVAL

Level	Signature	Date
Biometric:	_____	_____
Regional:	_____	_____
Divisional:	_____	_____

I. TITLE: Prince William Sound Herring Catch Sampling

II. OBJECTIVES:

A. List the specific objectives beginning with the highest priority:

1. Estimate the age and sex composition of the commercial catch of herring within the temporal and spatial strata sampled in each fishery.

2. Estimate the age and sex composition of the escapement. Use commercial purse seine catch samples or purse seine test fishery samples to represent spawning fish which escape commercial harvest.

3. Estimate the mean length, weight and fecundity by age and sex for each fishery and test fish catch sampled.

B. This project will contribute to the following Fisheries Management Operational Plans:

Species	Gear	Location
Herring	Purse Seine Sac Roe	Prince William Sound
Herring	Gill net Sac Roe	Prince William Sound

Herring	Pound Roe-on-kelp	Prince William Sound
Herring	Wild Roe-on-kelp	Prince William Sound
Herring	Purse Seine Bait	Prince William Sound

III. NEED OR PROBLEM ADDRESSED:

A. Describe the public and/or resource need addressed by the project and the project's benefits.

The Pacific herring stock in PWS is managed for a maximum exploitation of 20%. Timing of each fishery is established to maximize value; the spring roe fisheries to maximizing roe recovery; the fall bait/food fishery to optimize oil content and flesh quality. Harvest levels in all fisheries are forecast prior to the season based on the prior year's unharvested biomass and adjusted inseason based on the observed biomass and recruitment at age. Therefore the need to know the age composition of the spawning biomass present and any indication of recruitment of young ages is extremely important for the assessment of present and future biomass of PWS herring. Underlying the estimation of most components of a forecast (natural mortality, growth, recruitment, and escapement) is biomass (catch or escapement) at age. In addition, the processing industry has a great deal of interest in sex composition and average size data for predicting market value and roe recovery. Early in the season they use the data as an

indicator of potential roe recovery and post season as a cross check of roe recovery data, sex and size data provided by their own technicians inseason.

B. The success of the project will be judged by:

1. Success will be judged by the degree to which all sample collection goals are met, timeliness of inseason data analysis, and reporting.

2. By estimating the necessary Pacific herring population parameters, accuracy of the pre-season projection will increase and ability to accurately access run strength and distribution will increase. Success could be judged by the accuracy of the pre-season projection.

3. Success is also measured by whether results from the project are published in a timely fashion.

IV. PROJECT DESCRIPTION:

This project funds one Fisheries Biologist I and two Fishery Technician I's stationed in Cordova. Duties include processing age, sex, size, and fecundity samples collected from test fishing and pound monitoring (project TF-300) and from the commercial sac roe catch. The PWS Research Biologist (Sam Sharr PCN 1210) supervises

sample collection from onboard the R/V Montague. The Assistant Research Project Leader (Drew Crawford PCN 1337) supervises sample processing and preliminary analysis at the processing laboratory in Cordova. This is a basic biological sampling project designed to process time and area specific scale, sex, size, and fecundity samples from all of the PWS herring fisheries and spawning escapements. The data from this project sampling are used to provide accurate and timely estimates of the age, sex and size composition of PWS herring catches and spawning populations. In addition, temporal and spatial trends in the age composition of herring populations are identified. In conjunction with available biomass estimates, project data provide a tool for forecasting the future biomass of the herring population and, in conjunction with the new PWS herring spawn deposition project (project CF-303), provide accurate estimates of the fecundity at age for major spawning populations of herring for estimating the total spawning biomass in the area.

IV. A. Location:

Prince William Sound

B. Field Program Duration:

The spring sac roe and roe-on-kelp fisheries occur in April and frequently continue into the first week of May. The fall bait/food fishery was formerly from September through December but

now does not open until 1 November and the quota is usually harvested within a week to ten days. Three technicians are employed full time during the last three weeks of April to process samples from the roe fisheries. One of these technicians is retained for an additional week to help process any remaining samples, archive data, and dismantle the sampling laboratory. During the fall bait/food fishery two technicians are employed for one week each during the peak of the fishery in the first week of November.

C. Sampling Duration If Different Than Above: Samples from each fishery are flown directly to the Cordova area office as soon as possible after they are collected. Samples are processed on the same day or on the day following their arrival in Cordova. Typically five or six samples are collected per week during the spring roe fisheries and the sampling crew works full time. Approximately one day of each week is spent preparing for samples (i.e. label making, logging in samples, etc). Actual laboratory sampling takes approximately two and one half days. The remainder of the week is spent aging scales, preparing sample summary tables, and archiving sample data. The fall bait/food fishery now occurs over a short period; sampling is restricted to one stratum; the sample is processed immediately; and sampling efforts are completed within one work week.

D. Frequency Of Sampling While In The Field:

In the brief sac roe fisheries catches from each opening are sampled. If the catches are distributed over a large or discontinuous areas the catch sample is stratified by discrete bays or areas. Escapement samples are taken from any large area of spawn concentrations throughout the sound and at periodic intervals throughout the major spawning period (April). In the case of the pound fishery which usually occurs over an extended period, the sampling is stratified into early, middle, and late strata. In recent years the bait/food fishery has harvested the quota very rapidly. The fishery now opens on 1 November and the quota is reached within on week. One sample is taken when approximately half of the quota has been harvested.

E. Longevity Of The Project: [ ] 1 year, [ ] 2 years, [ ] 3 years, [X] continuing

F. Is this project new? [ ] Yes, [X] No

G. The project began or will begin 04/01/84

H. Give the title and status of the most recent project report.

Age, sex, and size composition of Pacific herring sampled from the Prince William Sound management area, 1984-1987, by Gene J. Sandone, published as an RIR 2A88-08.

Forecast of the Pacific Herring Biomass in Prince William Sound 1989, by Linda K. Brannian, published as an RIR 2A89-01.

V. DATA COLLECTION:

A. List types of data collected and means of recording each: Within each sample strata the data for each fish constitutes one record. One scale is collected from each fish and the following biological data are recorded: sex, length (in mm), weight (in gms), and age. In addition for each fish the following sampling data are recorded: date of capture, fishing district and subdistrict (from finfish statistical charts used for salmon catch reporting in PWS), local name for the location fished, name of the fishing vessel, the data form number and the fish number for that form. Scales are cleaned and mounted on glass slides for aging and permanent storage. All other biological and sampling data are entered into a microcomputer using a macro driven LOTUS spreadsheet and saved on diskette.

Herring AWL samples from major spawning sites in PWS are also subsampled for fish fecundity data which is analyzed and reported by the PWS Herring Spawn Deposition Study (Yellow Book Project No. CF-303). Each subsample consists of ten female fish in each 10 mm standard length interval in the 140 to 260 mm range. The ovarian sacs from each fish are bagged individually in zip-lock bags. A label is enclosed in each bag which includes the following

information: location where the fish was captured, data of capture, the length interval, and a reference to the AWL form number and fish number for the fish when sampled for scale, sex, and size data. Roe sac samples are then frozen for latter processing.

B. Sample Collection Methods: With the exception of a few minor changes the sampling methods used are described thoroughly in the PWS Herring Sampling Operational Plan (Sharr and Crawford, see attached) and the PWS Herring AWL Sampling Manual (Crawford, see attached).

VI. DATA ANALYSIS:

A. State sample sizes and how they were determined.

The sample size per time/area/gear stratum for age, length, weight, and sex data was set to simultaneously estimate proportions by age when sampling from a multinomial population. The goal was to select the smallest sample size for a random sample from a multinomial population such that the probability will be at least $1-\alpha$ (precision) that all of the estimated age proportions will simultaneously be within 5 percentage points (accuracy = 0.05) of the true population age proportions. The largest sample size for $\alpha = 0.1$ occurs when there are only 3 age classes present in equal proportions and guarantees at least this level of precision and accuracy for any number of age classes and proportions. This worst

case sample size[1] of 403 ageable scales will be used to estimate the age composition with the desired level of precision and accuracy in the absence of a better estimate of population proportions. Sample size for age, sex, and size sampling will be increased to 450 fish to account for unreadable scales.

Inseason a subsample of 200 fish may be aged and the proportions by age used as population parameters to estimate the subsequent sample size for that time/area/gear/fishery stratum. In this case higher precision (α = 0.05) will be sought. For example if the forecasted age composition is a good estimate of the true age proportions in 1989 a sample size of only 205 aged scales would be needed for our desired level of accuracy (0.05) and precision (α=0.05).

Sampling will be stratified by time, area, and fishery. Each fishing period will be a stratum. The pound fishery will be sampled early, middle and late in time. Further stratification may be necessary if gear is spread across a wide area. The goal for catch sampling is to estimate the age composition by fishing period, fishery, and area (if age composition varies by area for a given fishing period). The goal for escapement sampling is to sample each

[1] Thompson, S.K. 1987. Sample size for estimating multinomial proportions. The American Statistician 41:42-46.

peak biomass buildup in an aerial survey area defined by Brady (1987)[2].

B. List the types of data tables which you will use to summarize your data.

1. Age composition tables which show for each sex and for the sexes combined the following: the age specific portion of the sample in numbers of fish and as percent, the age specific mean length and weight of herring sampled, and the standard deviations of those means (see attached PWS Herring Sampling Manual, Table 3.).

C. State the types of statistical techniques and tests you expect to apply, and list the questions each test will help you evaluate.

To expose temporal and spatial trends in the age, sex, and size composition in the PWS herring sac roe fisheries test fishing samples taken almost daily prior to and after the fishery and catch samples during the fishery are collected from several proximate but discrete bays in the area encompassed by the fishery. Subsamples from each discrete sample are processed. Chi square tests on the

[2] Brady, J.A. 1987. Distribution, timing and relative biomass indices for Pacific herring as determined by aerial surveys in Prince William Sound 1978 to 1987. PWS D.R. 87-14.

age composition results from these subsamples expose differences between samples. If these subsamples from several temporally and spatially proximate samples are not significantly different they may be pooled into one sample. In this manner, the need to process many entire 450 fish samples is eliminated.

D. Specify the estimates (statistics) which are computed.

1. The sex and age composition of each sample is estimated as proportion of total sample by age class 2-13.

2. The mean length and mean weight by sex and age and the standard deviations around those means.

3. Use current and historic catch and population at age data to estimate mortality, stock size at age, stock size across years, recruitment at age, and average natural mortality at age across years.

E. Where, how, when, and with what hardware and software these analyses will be conducted. The age, sex and size analyses are done using a Lotus speadsheet data table which accesses data keypunched during the sampling and aging process. The forecast is done with a LOTUS spreadsheet macro built by the Region II and Headquarters commercial fisheries division Herring Research Staff.

VII. REPORTING:

A. Types of documents to be written by author and completion date.

Report	Author	Completion Date
PWS Herring C&E Leaflet (ADFG Tech. Fish. Rpt)	Brannian, Sharr & Crawford	Nov. 89
PWS Herring Forecast (ADFG Reg. Info. Rpt)	Brannian	Dec. 89
Appendix to Statewide Herring Forecast	Brannian	Nov. 89

VIII. PROJECT BUDGET:

A. By Line Item:

Line	GF	Other	Total
100	$ 9,700	_____	$ 9,700
200	$ 0	_____	$ 0
300	$ 100	_____	$ 100

400	$ 300	_____	$ 300
500	$ 0	_____	$ 0
Total	$ 10,100	_____	$ 10,100

B. The cost per sample for each data type.

Data Type	Cost/Observation
1.Age, sex, and size	$550 / 450 fish sample

C. Project Positions:

Class	PCN	PFT_mm	SFT_mm
FB III	1210	(Funded by CF-366)	
FB II	1337	(Funded by CF-366)	
FB I	1909		1.0
FT I	1515		1.0
FT I	1527		1.0

VIII. D. Man months assigned to each position for data analysis.

PCN	Report	mm
1210	Tech. Fish. Rpt. (C&E Leaflet)	1.0
1337	Tech. Fish. Rpt. (C&E Leaflet)	1.0
1100	Tech. Fish. Rpt. (C&E Leaflet)	0.2
1100	Reg. Info. Rpt. (Forecast)	0.2

E. Man months assigned to each position for report writing and other presentations of project data?

PCN	Report	mm
1210	Tech. Fish. Rpt. (C&E Leaflet)	0.5
1337	Tech. Fish. Rpt. (C&E Leaflet)	0.5
1100	Tech. Fish. Rpt. (C&E Leaflet)	0.5
1100	Reg. Info. Rpt. (Forecast)	0.3
1210	Board of Fisheries Reports	0.3
1337	Board of Fisheries Reports	0.3

APPENDIX D.2

PRINCE WILLIAM SOUND HERRING A-W-L SAMPLING MANUAL

Alaska Department of Fish and Game
Commercial Fisheries Division
Box 669
Cordova, Alaska 99574

February 1989

APPENDIX F

Laboratory Studies of Oil Impacts on Pacific Herring Eggs and Larvae

Proposal for Professional Services

Prepared for:
Minerals Management Service
949 East 36th Avenue, Room 110
Anchorage, Alaska 99508-4302

Prepared by:
ENVIROCON PACIFIC LIMITED
#205-2250 Boundary Road
Burnaby, British Columbia
Canada V5M 3Z3

tel: (604) 291-0292
fax: (604) 281-2139

April 1989

1.0 Introduction

The study plan of this proposal was developed in cooperation with biologists of the Commercial Fish Division of the Alaska Department of Fish and Game (ADFG), particularly Mr. Fritz Funk and Ms. Evelyn Biggs, and with Dr. Jeffrey Marliave of the Vancouver Public Aquarium. The plan is only one part of a 7-part plan to assess the impact of the recent Exxon Valdez oil spill on the stock of herring that spawns in Prince William Sound. A list of the 7-part plan is enclosed in this proposal.

The 7-part herring impact study includes 3 parts that deal with eggs and larvae:

(5) field studies of the mortality of eggs laid on oiled and non-oiled spawning beaches in Prince William Sound;

(6) laboratory studies of the percentage hatch of viable larvae from herring eggs taken from oiled and non-oiled beaches in the Sound; and

(7) field studies of the growth and fitness of herring larvae captured in oiled and non-oiled areas of the Sound.

Part (5) is designed to measure the percentage of herring eggs that die on oil-impacted beaches and compare this with the percent mortality of eggs on pristine spawning beaches. It is based on SCUBA surveys of the ratio of live to dead eggs on several separate spawning beaches. These surveys will be carried out by ADFG personnel; they are not the subject of this proposal.

The SCUBA surveys will assess egg mortality, but they cannot provide information on the fate of the very young larvae that hatch from eggs that survive the 20 d incubation period. This information can only be obtained by incubating small batches of eggs from the same beaches that are surveyed by SCUBA in laboratory aquaria, capturing the larvae that hatch, examining them for visible abnormalities, and testing their response to food. These incubation experiments [part (6)] are the subject of this proposal.

The field study of larval growth and fitness [part (7)] is described in a separate proposal.

2.0 Objectives

The primary objective of this study is to describe the relationship(s) between percent hatch of viable herring larvae and the degree of oil contamination of the spawning beaches. The secondary objective is to present the results of this study in a form that allows ADFG to calculate the proportion of live eggs on specific spawning beaches in Prince William Sound that produced viable larvae.

3.0 Study Plan

3.1 Experimental Design

The design of the experiments is based on a 2-way mixed model analysis of variance (ANOVA)

$$Y_{ijkl} = u + a_i + b_j + C_k + (ab)_{ij} + e_{ijkl}$$

where

Y_{ijkl} = an observation of the lth replicate egg batch of the jth depth class, of the kth transect, of the ith oil treatment;

"Observation" is the: ratio of live to dead eggs, rate of egg mortality over the incubation period, percent total hatch, percent of larvae with visible abnormalities, mean developmental stage, mean length at hatch, mean yolk sac volume, mean dry weight, mean condition factor (weight length^{-3}), and other parameters.

u = grand mean;

a_i = fixed deviation from u caused by beach treatment (non-oiled, light-oil, medium-oil, and heavy-oil);

b_j = fixed deviation from u caused by depth class (intertidal, mid-water, subtidal) from which the eggs were sampled;

C_k = random deviation from u caused by the grouping of depth classes into transects;

$(ab)_{ij}$ = first-order interaction between beach treatment and depth class; and

e_{ijkl} = unexplained error variance.

The design is shown in Table 1. This design will allow the testing of the significance of a_i, the beach treatment, and $(ab)_{ij}$, the interaction of beach treatment and water depth.

ABLE 1
Experimental design for laboratory incubation of herring eggs

	BEACH TREATMENT																			
	Control					Low-oil					Medium-oil					High-oil				
	transect					transect					transect					transect				
WATER DEPTH	1	2	3	4	5	1	2	3	4	5	1	2	3	4	5	1	2	3	4	5
Low	3	3	3	3	3	15					15					15				
Medium	3	3	3	3	3	15					15					15				
High	3	3	3	3	3	15					15					15				
Samples no:	45					45					45					45				= 180

Notes:
1. 3 replicate batches of eggs are in each of the 3 cells of each of the 20 transects (4 treatments x 5 transects each), giving 3 x 3 x 20 = 180 batches of eggs.

2. The cells in the 3 oiled beach treatments are not shown for the sake of clarity.

3.2 Egg Collection and Transport

Batches of about 300 live eggs will be collected by ADFG divers as part of their survey of live/dead egg ratios on the spawning beaches of Prince William Sound. Each batch will be placed in a wet, porous paper bag to which is securely fasened a label giving the beach, transect, depth of collection, replicate number, date and time of collection, and name of sampler. Then each egg batch will be packed in an insulated travelling chest between layers of crushed sea ice and wet paper. A pan will be placed at the bottom of each chest to collect melted seawater. This pan will prevent eggs and melted water from mixing and so prevent the eggs from asphyxiating in the water. This method can keep herring eggs alive and healthy for up to 24 hours after collection.

The chests will be flown by ADFG to Cordova within 6 hours of collection. An Envirocon Pacific employee will transport the chests from the plane to the airport and place them on the 1600 hour flight to Seattle (5 hours). There the eggs will either be flown to Vancouver (1 to 2 hours) or collected at the airport and driven to Vancouver (2.5 hours by car). The total elapsed time from collection to delivery to the laboratory will be 14 to 20 hours.

Enviroconn Pacific Limited will send a trained technician, and at least 6 insulated chests, to Cordova at least 1 day before the beginning of ADFG's spawn survey. His duties will be to: (1) instruct the ADFG surveyors on how to pack the eggs in the chests, perhaps by accompanying them on their first sampling date; (2) pick up each chest as soon as it arrives in Cordova's harbor, drive it to the airport, and ensure that the eggs are placed on the next flight to Seattle; and (3) arrange for the collection of new samples in the event that bad weather or accidents prevent the timely delivery of the eggs to Cordova or Seattle.

3.3 Egg Incubation

Upon delivery to the laboratory of the Vancouver Public Aquarium, each egg batch will be placed in clean cool seawater in its own labelled glass or plastic container. Gentle aeration will be provided by compressed air pumped through an airstone. The containers will be cooled to about 8°C in a large water bath.

Each day the following variables will be measured from each batch:
(1) ratio of live eggs to dead eggs (dead eggs are opaque, live eggs are clear);
(2) number of newly-hatched larvae;
(3) percent of larvae with visible abnormalities, e.g. missing lower jaw, flexed spine, etc.;
(4) average developmental stage of the larvae upon hatch (using Doyle's classification scheme for Atlantic herring larvae);
(5) average yolk sac volume (an index of prematurity);
(6) average length;
(7) average dry weight; and
(8) average condition factor (dry weight length^{-3}).

Newly-hatched larvae will be preserved in 5% formalin on the day of capture for later microscopic examination and possible histological examination, except for a subsample of about 10 larvae which will be kept live in a separate container for feeding experiments. These experiments involve exposing groups of 2 to 5 d old herring larvae to 1 d old brine shrimp nauplii in a glass dish for 1 hour, and then counting the average number of nauplii visible through the gut wall of the herring larvae. Herring larvae do not begin to feed until day 2, and they completely absorb their yolk sac by day 5 to 6.

.0 Fate of Eggs and Larvae

s stated in section 3.0, all larvae will be killed in formalin oon after hatch. Dead eggs and the vegetation to which they re attached will be disposed of as biological waste. The water sed to incubate the eggs will be disinfected with bleach and oured into municipal drains. The staff of the Vancouver Public quarium is aware of the necessary procedures.

5.0 Schedule

ADFG proposes to begin spawn surveys in Prince William Sound on April 20, 1989. Eggs for the incubation study should be collected as early in the survey as possible, so that additional samples can be collected at a later date, if required. Since eggs will incubate for about 20 days, the laboratory part of the study will be completed by May 31, 1989. A final report will be submitted by January 1, 1990.

PREFACE

The following document is a revised version of the "Bering Sea Herring A-W-L Sampling Manual, 1987" that was written by Robert Lebida for the Bristol Bay and A-Y-K areas. Revisions and additions were made in order to document the herring age, weight, and length (AWL) sampling procedures that are currently being used in Prince William Sound (PWS). Many of the procedures that are discussed in this manual were initiated by Sam Sharr, the Research Project Leader for the Commercial Fisheries Division in the Prince William Sound area. The documentation of these PWS herring sampling procedures was done by Drew Crawford, the Assistant Research Project Leader in Cordova.

Herring AWL samples in Prince William Sound are collected by a sampling crew aboard the R/V Montague and by designated fishing vessels in the commercial purse seine fleet. Clearly labeled samples in polyethylene bags are picked up daily or as weather permits by float planes that are chartered by department personnel to fly aerial surveys of Prince William Sound during the herring season. All PWS herring samples are flown to Cordova, delivered to the lab at the Cordova ADF&G office, placed in fish totes, iced down with snow, and are logged in a sampling log. All samples are then worked up in the warm, dry, well-light surroundings of the Cordova office lab.

This type of a sampling environment has enabled us to do away with the standard mark-sense AWL forms and enter the data directly into a LOTUS 1-2-3 spreadsheet on a Compaq Plus computer. This approach eliminates several data processing steps (e.g. logging mark-sense data forms, editing mark-sense data forms, shipping mark-sense data forms to be OPSCANed, waiting for OPSCAN results to be shipped back, and correcting OPSCAN data printouts). Macro functions in the data entry spreadsheet also enable us to access the data and produce summary tables of the results soon after the data has been entered. Therefore, we have shortened our turn-around time considerably and have even been able to provide same-day AWL sample results to fishery managers when the samples are received by mid-morning.

Individuals who are familiar with LOTUS 1-2-3 and computer procedures may find the documentation of computer procedures in this manual to be excessive. To those individuals I apologize for my cook book approach. However for those individuals who are not familiar with LOTUS 1-2-3 and have had little or no computer training, I hope that I have documented these procedures well enough so that you can accomplished these tasks with a minimum of frustration.

This procedures manual will be reviewed annually and revised and updated as needed. If you have comments or suggestions for improving any of the methods or procedures that are discussed in this draft, please contact Drew Crawford at the ADF&G office in Cordova (424-3212).

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INTRODUCTION

Herring from commercial and test purse seine and gill net catches are sampled for sex, size, and age annually in Prince William Sound to form a data base essential to the management of the herring resources. This information is drawn upon by management and research biologists for monitoring and regulating harvest levels, determining run timing, entry patterns, and distribution of herring arriving on the spawning grounds, monitoring sexual maturity and age composition of herring spawning populations, developing methods to forecast herring abundance and determine optimum spawning goals, and to gain a better understanding of the biology of each stock. The usefulness of this AWL data depends on the manner and accuracy in which the samples were taken. This manual was prepared as a guideline for collecting information on age, sex, length, and weight of herring. The sampling and recording procedures are listed in a logical order of activity and should be followed in sequence to develop accurate sampling techniques.

EQUIPMENT LIST

- 4 - Fish totes, small gray plastic (11.5" X 18.5" X 31.5")
- 1 - Fish tote, large white fiberglass wi/ lid (25" X 37.5" X 25")
- 2 - Sample basins, white plastic (8" X 15" X 16")
- 6 - Sample trays, yellow (1/4" X 11" X 24")
- 1 - Light table, Porta-Trace (Model No. 1618-4)
- 120 - Glass microscope slides, plain, clear, (25 X 75 X 1 mm)
- 2 - Scotch tape (0.5 - 1.0 inch [13-25 mm] wide)
- 2 - Forceps
- 2 - Scissors, paper cutting
- 1 - Scissors, dissecting (used for obtaining fecundity samples)
- 2 - Scalpels wi/spare blades
- 3 - Beakers, 100 ml plastic wi/ lids
- 2 - Dishes, plastic (1/4" X 2" X 2") for 1:10 mucilage glue solution
- 1 - Caliper, Mitutoyo (30 cm)
- 1 - Scale, Ohaus (Brainweigh B300)
- 60 - Sample labels (computer generated)
- 60 - Slide labels (computer generated)
- 1 - Paper towels (roll)
- 1 - Mucilage glue (3 oz.)
  Water
- 1 - Compaq Plus computer
- 2 - Diskettes, preformatted and labeled (Original and Backup)
- 1 - 1988 PWS Herring AWL Data and Summary Table Notebook (black, 3-R binder) with program files and protective plastic sleeves for filing and storing data diskettes:
  a. HTAB.WK1 - LOTUS 1-2-3 macro file which generates sample labels used on yellow sample trays.
  b. SLIDEL.WK1 - LOTUS 1-2-3 macro file which generates slide labels.
  c. HAWLV288.WK1 - LOTUS 1-2-3 macro file for herring A-W-L data entry and generating summary tables
- 1 - Microfiche reader

METHODS

I. Preparation Prior to Data Collection

A. Before any fish are handled, make up the sample and slide labels using the Compaq Deskpro computer and the Okidata printer in Drew's office. The program diskettes for making these labels are located in the front section of the black 3-ring binder entitled "1988 PWS Herring AWL Data and Summary Tables". This notebook will be kept in the lab by the computer during the herring season. All herring labels are produced with LOTUS 1-2-3 Macro Files.

1. Sample Labels - Use File Named: HTAB.WK1

a. Type the "highlighted" response to the computers prompts and depress the [RETURN] key after each entry.

```
C:\>
C:\>CD\123    (Changes to the LOTUS 123 subdirectory)
C:\123>123    (Initiates the LOTUS 123 program and displays
                    a blank spreadsheet on the screen)
```

Insert the diskette that contains the HTAB.WK1 file in the A-Drive.

To retrieve this file for use, do the following:

Press slash "/" key (Displays the 1-2-3 Command Menus)

Depress the first letter of the following commands in the order that is indicated - FILE, RETRIEVE

The computer will highlight the first file on your diskette which is HTAB.WK1. To call up this file, press the [RETURN] key.

The computer will now display the last labels that were SAVED on the HTAB.WK1 file. An example of a typical SAMPLE LABEL that is produced by this file is shown below:

```
WELLS BAY TEST P/S SAC ROE FISHERY 4/10/88
     WELLS BAY TEST P/S SAC ROE FISHERY 4/10/88   001A
     WELLS BAY TEST P/S SAC ROE FISHERY 4/10/88   001B
     WELLS BAY TEST P/S SAC ROE FISHERY 4/10/88   001C
     WELLS BAY TEST P/S SAC ROE FISHERY 4/10/88   002A
     WELLS BAY TEST P/S SAC ROE FISHERY 4/10/88   002B
     WELLS BAY TEST P/S SAC ROE FISHERY 4/10/88   002C
     WELLS BAY TEST P/S SAC ROE FISHERY 4/10/88   003A
       .    .    .    .    .    .    .    .    .
       .    .    .    .    .    .    .    .    .
       .    .    .    .    .    .    .    .    .
     WELLS BAY TEST P/S SAC ROE FISHERY 4/10/88   020C
```

b. Enter the new label information on the top line only (e.g. Location, Sample Type, Gear Type, Fishery, and Date [mm/dd/yy]), then press [RETURN].

c. The other lines are programed to change automatically after you enter the new information on the first line.

d. Print out the sample labels on the Okidata printer on 8 1/2 x 11 inch paper by doing the following:

Press the slash "/" key. (Displays the 1-2-3 Command Menus)

Depress the first letter of the following commands in the order that is indicated -

PRINT
PRINTER [RETURN]
RANGE (Preset for A2..G62) [RETURN]
ALIGN (Before you press GO, turn on the Okidata printer and make sure that it is loaded with 8 1/2 X 11 inch paper)
GO (The new labels will be printed out by the printer)

e. Remove the printout of sample labels from the printer and make a cut between each sample label from the right margin toward the left margin with a pair of scissors. Stop each cut about 1/4 to 3/8 inches from the left margin. By leaving the left margin in tact, the labels will stay in order, but can easily be torn off one-by-one as they are needed.

2. Slide Labels - Use File Named: SLIDEL.WK1

a. Type the "highlighted" response to the computers prompts and depress the [RETURN] key after each entry.

C:\>
C:\>CD\123 (Changes to the LOTUS 123 subdirectory)
C:\123>123 (Initiates the LOTUS 123 program and displays a blank spreadsheet on the screen)

Insert the diskette that contains the SLIDEL.WK1 file in the A-Drive.

To retrieve this file for use, do the following:

Press slash "/" key (Displays the 1-2-3 Command Menus)

Depress the first letter of the following commands in the order that is indicated - FILE, RETRIEVE

The computer will highlight the first file on your diskette which is HTAB.WK1. Press the right arrow ( ) cursor key once. The computer will highlight the second file on the diskette which is named SLIDEL.WK1. To call up this file, press the [RETURN] key.

The computer will now display the last labels that were SAVED on the SLIDEL.WK1 file. An example of a typical SLIDE LABEL that is produced by this file is shown below:

```
FISH NO. 1          5    11          15    21          25
   SLIDE NO.        SLIDE  0A        SLIDE  0B        SLIDE  0C
   SPECIES          HERRING          HERRING          HERRING
   LOCATION         GALENA BAY       GALENA BAY       GALENA BAY
   SMPL TYPE        SAC ROE TEST     SAC ROE TEST     SAC ROE TEST
   DATE             08 APR 1988      08 APR 1988      08 APR 1988
   FISH NO.       6           10   16          20   26          30

        1           5    11          15    21          25
                    SLIDE  1A        SLIDE  1B        SLIDE  1C
                    HERRING          HERRING          HERRING
                    GALENA BAY       GALENA BAY       GALENA BAY
                    SAC ROE TEST     SAC ROE TEST     SAC ROE TEST
                    08 APR 1988      08 APR 1988      08 APR 1988
                  6           10   16          20   26          30

                  1           5    11          15   21          25
                    SLIDE  2A        SLIDE  2B        SLIDE  2C
                    HERRING          HERRING          HERRING
                    GALENA BAY       GALENA BAY       GALENA BAY
                    SAC ROE TEST     SAC ROE TEST     SAC ROE TEST
                    08 APR 1988      08 APR 1988      08 APR 1988
                  6           10   16          20   26          30

                  1           5    11          15   21          25
                    SLIDE  3A        SLIDE  3B        SLIDE  3C
                    HERRING          HERRING          HERRING
                    GALENA BAY       GALENA BAY       GALENA BAY
                    SAC ROE TEST     SAC ROE TEST     SAC ROE TEST
                    08 APR 1988      08 APR 1988      08 APR 1988
                  6           10   16          20   26          30
                      .                .                .
                      .                .                .
                      .                .                .

                  1           5    11          15   21          25
                    SLIDE 20A        SLIDE 20B        SLIDE 20C
                    HERRING          HERRING          HERRING
                    GALENA BAY       GALENA BAY       GALENA BAY
                    SAC ROE TEST     SAC ROE TEST     SAC ROE TEST
                    08 APR 1988      08 APR 1988      08 APR 1988
                  6           10   16          20   26          30
```

b. Press the "Caps Lock" key, so that all new entries will be typed as CAPITAL letters.

c. Use the up ( ) and down ( ) cursor keys to move the highlighted cell to the category that you wish to change within the data entry cell (C4..C8).

d. Type in the new information for each category (e.g. CARD, SPECIES, LOCATION, SAMPLE TYPE, DATE [dd/mm/yy]) that needs to be changed, and press [RETURN] after each new entry.

Note: The other labels on the screen will be automatically be updated with the new information in each category, each time you press the [RETURN] key.

For a new sample, the only slide label categories that will need to be changed are LOCATION, SAMPLE TYPE, AND DATE. For the second, third, or fourth sample from the same location with the same sample type, the scale SLIDE NUMBER will also have to be changed to keep these samples in sequential order.

(e.g. Sample 1 - Wells Bay Sac Roe Test 08 Apr 1988, Slide Nos. 1A-20C;
Sample 2 - Wells Bay Sac Roe Test 15 Apr 1988, Slide Nos. 21A-40C;
Sample 3 - Wells Bay Sac Roe Test 24 Apr 1988, Slide Nos. 41A-60C).

e. Print out the slide labels on the Okidata printer on 8 1/2 x 11 inch paper by doing the following:

Press slash "/" key (Displays the 1-2-3 Command Menus)

Depress the first letter of the following commands in the order that is indicated -

PRINT
PRINTER [RETURN]
RANGE (Preset for B12..V193) [RETURN]
ALIGN (Before you press GO, turn on the Okidata printer and make sure that it is loaded with 8 1/2 X 11 inch paper)
GO (The new labels will be printed out by the printer)

f. Reduce the labels 64% on the copy machine.

g. Cut out each slide label with a scissor and glue one slide label on the left side of each slide with 1:10 solution of mucilage glue and water.

B. Miscellaneous Preparations:

1. Refer to the Prince William Sound finfish statistical area chart (Figure 1) to determine the location code for the sample.

2. Fill one beaker with water and one beaker with a solution of 10 parts water to 1 part mucilage glue.

3. One gross or two boxes of microscope slides are needed for each Prince William Sound herring sample (n=450 fish). The supply of microscope slides is located in the back of the bottom file drawer next to the scale press in Drew's office.

   a. Ten scales will be mounted on each slide (one scale from each of 10 herring).
   b. A second slide will be used to cover the first after they are filled with scales.
   c. Three complete scale mounts (e.g. 10 scales sandwiched between two slides) correspond to fish on one A-W-L form (e.g. A-W-L #001 = Slides 1A, 1B, & 1C).

4. A set of three glass slides and their corresponding A-W-L data should not be used for scales or data for more than one day, one district, one subdistrict, one gear type, or one mesh size.

5. The slides and corresponding A-W-L data are numbered sequentially by date throughout the season starting with 1A, 1B, 1C, 2A, 2B, 2C, 3A, 3B, 3C, etc.

   a. A separate numbering sequence beginning with 1A will be used for each district, subdistrict, and gear type (e.g. Eastern District, Olsen Bay, Test P/S, 1A-20C; Northern District, Unakwik Inlet, Comm Sac Roe G/N, 1A-20C).
   b. Keep dates and page numbers in proper sequence.

II. Measurements and Observations

A. <u>Weight Data Collection</u>:

1. Pick out ten herring from the sampling tote. Keep in mind that fish to be sampled should be selected "RANDOMLY". One way of doing this is to reach down into the sampling tote and select the first fish that you grab on to. Also make an effort to select fish from different areas and depths of the tote. Place each herring flat on its right side on a yellow sampling tray in vertical rows of 10 fish each. Fish number "1" is on the top of the sampling tray and fish number "10" is on the bottom. Tear off a sample label (e.g. Olsen Bay, Test P/S, Mar 30, 1988, Sample 001A) and place it on the tray between the fifth and sixth fish.

2. Starting with the top fish in the row, weigh each herring on the sampling tray to the nearest gram using a Dial-O-Gram type scale.

3. Call out the weight measurement to the nearest gram so that the information can be entered by the keypuncher into the Compaq Plus computer.

4. Follow the same procedure for measuring the weights of the remaining fish in that group of ten.

B. Standard Length Data Collection:

1. Then start with the top fish on the sampling tray again and measure the standard length of each fish with calipers (Figure 2). Place the left end of the calipers on the tip of the lower jaw and slide the right end of the caliper down the lateral line of the fish until you get to the crease where the hypural plate meets the caudal (tail) fin rays.

2. Call out the length measurement to the nearest mm so that the information can be entered by the keypuncher into the Compaq Plus computer.

3. Follow the same procedure for measuring the lengths of the remaining fish in that group of ten.

C. Sex Data Collection:

1. Sex is initially checked by milking the gonads of each fish to produce evidence of milt (males) or eggs (females) at the vent. If no sex product is observed, then an incision is made in the abdomen of the fish with a scalpel to determine the sex by examining the gonads. Numerical sex codes are given in Table 1.

2. Call out the sex so that the data can be keypunched in the computer.

D. Fecundity Data Collection:

Herring AWL samples in Prince William Sound are subsampled in select areas to obtain fish for fecundity enumeration. Five separate samples are collected form AWL samples from five areas. Ten female fish are sampled in each 10 mm length interval from 140 to 260 mm standard length as available in each length class. The ovarian sacs from each sample are placed in a twirl-lock plastic bags with a label that identifies the sample. The label contains the following information: location where the fish was captured, date of capture, the length interval, and a reference to the AWL form number and fish number. The samples are then frozen for later processing. These samples are collected for the Prince William Sound Herring Spawn Deposition Study (Yellow Book Project No. CF-303) and the analysis of these data are presented in the Spawn Deposition Study Report.

III. Scale Sampling

A. Only one readable scale will be taken from each herring with a maximum of 10 scales placed on each slide (Figure 3.).

B. Remove each scale from one of the preferred body areas (Figure 4) on the left side of the fish (right side of the fish sampled as an alternate if necessary) using forceps. Body area locations are numbered in order of preference (Location 1 is most preferred, Location 5 is least preferred).

C. Dip each scale in clean water, rub between thumb and forefinger to remove dirt and slime, examine (hold up to a light) for regeneration (regenerated scales appear blurred in the center). DISCARD IF REGENERATED, repeat the procedure until a suitable scale is located.

D. To mount a scale on the glass slide, dip the scale into the mucilage glue solution (1 part mucilage glue : 10 parts of water), shake off excess solution, and place the scale onto the labeled glass slide. Make sure that the unsculptured (concave) side of the scale is facing down and the anterior margin (portion embedded in the integument of the fish) is facing towards the top of the slide. The ridges on the sculptured side of the scale can be felt with a fingernail or forceps. Make certain that the scales are placed on the slide that has the same sample number as the sample from the yellow tray and that the scales are placed on the slide in the positions that correspond to the correct specimen number in that sequence of 10 fish (Figure 3).

E. Press each scale firmly against the slide with a paper towel after mounting to remove excess glue from underneath the scale.

F. Place a second slide on top of each frosted slide when all scales have been mounted. Tape both slides together with a piece of Scotch tape at the labeled end only. Make certain tape does not cover any mounted scales.

G. After all 10 scales have been mounted on the slide, and before you discard the fish on the sampling tray in the disposal basket, double check the label on the slide with the label on the sample tray to make sure that they agree.

H. Store completed scale mounts in a slide box to avoid loss or breakage. Label the slide boxes according to year, fishery, location, sample type, gear type, dates, and slide numbers.

(e.g. 1988 PWS Sac Roe Herring,
Olsen Bay, Test P/S Sample, 4/31/88
001A-020C)

I. Enter the age of each specimen in the computer in the appropriate block as the fish are age with the microfiche reader. The age code for a

regenerated scale is 18, an illegible scales is 19, and a missing scales is 20 and wrong scale is 21.

IV. Data Entry

A. Since all Prince William Sound herring A-W-L samples are worked up in the warm, dry, well-light surroundings of the Cordova office lab, we have dispensed with the use of the traditional mark-sense A-W-L data forms and elected to enter the data directly into a LOTUS 1-2-3 spreadsheet on a Compaq Plus computer. This allows us to eliminate several data processing steps (e.g. logging mark-sense forms, editing mark-sense forms, shipping mark-sense forms to be OPSCANed, waiting for the OPSCAN results to be shipped back, and correcting data summary printout errors). It also enables us to access the data and produce summary tables immediately after all the A-W-L data has been entered.

B. The LOTUS 1-2-3 macro file which is used to enter and summarize herring A-W-L data is named HAWLV288.WK1. This file is located on a diskette on page 2 of the "1988 PWS Herring AWL Data and Summary Table Notebook".

1. Turn on the Compaq Plus computer and enter the LOTUS subdirectory.

   C:\>
   C:\>CD\LOTUS
   C:\LOTUS>LOTUS (The computer will highlight "1-2-3")

2. Access the 1-2-3 spreadsheet program by pressing the [RETURN] key. A blank 1-2-3 spreadsheet will be displayed on the monitor.

3. At present all of the AWL data entry and data summary files which are discussed in this procedures manual are on diskettes (original and backup copy for each). Therefore you must either preset your computer to operate off the A-Drive or do the following each time you turn on your computer to work with these files.

   To change to the file directory to the A-Drive do the following:

   Press the slash "/" key. (Displays the 1-2-3 Command Menus)

   Depress the first letter of the following commands in the order that is indicated - FILE, DIRECTORY (The computer will indicate the default directory)

   Enter current directory: C:\LOTUS

   To change the directory to the A-Drive, type in A:\ and press t h e [RETURN] key.

4. Insert the diskette which contains the LOTUS 1-2-3 macro file named HAWLV288.WK1 into the A-Drive. This diskette is labeled "Herring Sampling Data Entry Spreadsheet and Summary Macro's" and

is located on the second page of the "1988 PWS Herring AWL Data and Summary Table" 3-ring notebook in the lab.

5. To retrieve this file for use, do the following:

Press the slash "/" key. (Displays the 1-2-3 Command Menus)

Depress the first letter of the following commands in the order that is indicated - FILE, RETRIEVE (The computer will ask,

Name of file to retrieve: A:*.wk ?

and it will highlight the first file on the diskette which is HAWLV288.WK1. To call up this file, press the [RETURN] key.

The computer will display the upper left section of the HAWLV288.WK1 file on the monitor which is shown below:

```
B4: [W4] '(ALT)N - To name data file and enter header info for 1st AWL.  READY

     A    B    C    D    E    F    G    H    I    J    K  L   M    N    O
40    M    D    G   DI   SE   LC TIME  HRS  AWL#  F#  SX  GI  LEN  WT  AGE

1   This is a macro driven spreadsheet for herring AWL data entry and 2   analysis.
The following MACROS are available:
3
4          (ALT)N - To name data file and enter header info for 1st AWL.
5          (ALT)C - To update date file and enter header info for next AWL.
6          (ALT)E - To end data entry, save file, and clear the spreadsheet.
7          (ALT)P - To recalc. the sum. table and save it in wks & prn files.
8          (ALT)O - To resave an old data file which has been imported into the
9                   macro for aging and editing.
10
11
12
13
14
15
16
17
18
19
27-Apr-88  06:42 PM
```

The information in this section of the 1-2-3 spreadsheet (Range - A1..09) lists the macro options that are available in this file. Note - the (ALT)E and (ALT)O macro options were not used in 1988.

6. Creating an AWL Data File With the (Alt) N Macro:

After you have retrieved the HAWLV288.WK1 file you are ready to create a new herring AWL data file and beginning entering data. This can be done as follows:

IMPORTANT! First remove the diskette which contains the HAWLV288.WK1 file from the A-Drive and insert an empty preformatted data diskette. The HAWLV288.WK1 file is a master template which is only used to retrieve this LOTUS 1-2-3 macrofile. No data should be entered or saved on the diskette which contains the master copy of HAWLV288.WK1.

Data entry diskettes are located on subsequent pages in the "1988 PWS Herring AWL Data and Summary Table Notebook" and are arrange according to year, fishery, sample type, and gear type. Select the appropriate data entry diskette (e.g. 1988 PWS Herring Sac Roe Fishery Test Purse Seine Data) and proceed as follows:

a. Press the "Alt" key and the letter "N" key simultaneously.

This invokes the (ALT)N macro and enables you to name the data file, enter header information on the data entry form that will identify this AWL data set, and then begin entering data. The computer will then ask a series of questions about the sample that you wish to enter. Press the "Caps Lock" key. Then type in the appropriate response to each question and depress the [RETURN] key after each response.

NAME FOR DATA FILE (8 CHARACTERS)? (e.g. 88HTWBD1.WK1)

NOTE - In Prince William Sound we have establish the following convention for naming AWL data files for herring:

Year (e.g. 88),
Type of Sample (e.g. HT = Herring Test,
HC = Herring Commercial Catch, and
HP = Herring Pound Sample),
Location Where the Sample was Collected (e.g. WB = Wells Bay),
Type of File (e.g. D = Data, T = Summary Table),
Number in the Series (e.g. 1 = this is the first sample of this type from the indicated location with this gear type), and
File Extension (.WK1 is an ending that identifies all LOTUS 1-2-3 spreadsheet files).

ENTER A BRIEF DATA DESCRIPTION? (e.g. 1988 PWS Sac Roe Herring, Wells Bay Test P/S Sample, 4/10/88.)

NOTE - The Prince William Sound convention for describing or assigning a title to AWL data files for herring are:

Year (e.g. 88),
Management Area (PWS=Prince William Sound),
Fishery (e.g. Sac Roe Herring or Food/Bait Herring),
Location of Sample (e.g. Wells Bay),
Sample Type (e.g. Test, Com. Cat.= Commercial Catch, or Pound),
Gear Type (e.g. P/S = Purse Seine or G/N =Gill Net), and
Date (mm/dd/yy).

The computer will then display the data entry section of this LOTUS 1-2-3 spreadsheet (Range - A41..060) on the monitor and ask you to answer questions which will fill in the column header information. Answer the questions as indicated and press the [RETURN] key after each response.

A41: [W4]
SAMPLE MONTH (NN) ?

	A	B	C	D	E	F	G	H	I	J	K	L	M	N	O
40	M	D	G	DI	SE	LC	TIME	HRS	AWL#	F#	SX	GI	LEN	WT	AGE
41															
42															
43															
44															
45															
46															
47															
48															
49															
50															
51															
52															
53															
54															
55															
56															
57															
58															
59															

27-APR-88 07:01 PM CMD CALC CAPS

Questions that will be asked about the sample include the following:

SAMPLE MONTH (NN) ? (e.g. 04 = April)

SAMPLE DAY (NN) ? (e.g. 10)

NOTE - The sample day is the day the sample was collected, not the day the sample is worked up in the lab.

GEAR CODE (NN) ? (e.g. 05 = Purse Seine, 04 = Drift Gill Net)

NOTE - A complete listing of gear codes can be found in the upper left hand corner of Table 1.

DISTRICT CODE (NNN) ? (e.g. 222 = Northern District)

NOTE - District codes for the Prince William Sound Area can be found in Figure 1.

SECTION CODE (NN) ? (e.g. 20 = Section where Wells Bay is located)

NOTE - Section codes for the Prince William Sound Area can also be found in Figure 1.

LOCATION CODE (NN) ? This code is not used in Prince William Sound, press the [RETURN] key to bypass this question.

SAMPLE TIME (NN) ? (e.g. 08 = 0800 hours, military time) The sample time will not always be known but whenever it is enter it here.

NO. HRS. FISHED (NNN) ? This code is not used in Prince William Sound, press the [RETURN] key to bypass this question.

BEGINNING AWL # (NNN) ? (e.g. 001)

NOTE - For a new sample, the appropriate response to this question is 001. However, if this is the second, third, or fourth sample of the same type and gear type for a particular location, enter the next number in the series that would follow after your last sample.

For example if you had completed a 600 fish sample (AWL 001-020, 30 fish per AWL) of Test P/S herring for Wells Bay on April 10 and you

get a similar sample from Wells Bay on April 20, the second sample will start off with AWL 021.

HOW MANY FISH ON THIS CARD (NN) ? (e.g. 30)

NOTE - This data entry spreadsheet is set up just like a standard mark-sense AWL data form which allows 30 fish on each AWL data form. Therefore the appropriate response to this question will usually be 30 unless you are at the end of your sample and you don't have enough fish to complete a full AWL form.

At this point the (ALT)N macro will print out the appropriate header information for the samples 001-030 on AWL 001 as follows:

K41: [W3]

	A	B	C	D	E	F	G	H	I	J	K	L	M	N	O
40	M	D	G	DI	SE	LC	TIME	HRS	AWL#	F#	SX	GI	LEN	WT	AGE
41	4	22	5	222	20				1	1					
42	4	22	5	222	20				1	2					
43	4	22	5	222	20				1	3					
44	4	22	5	222	20				1	4					
45	4	22	5	222	20				1	5					
46	4	22	5	222	20				1	6					
47	4	22	5	222	20				1	7					
48	4	22	5	222	20				1	8					
49	4	22	5	222	20				1	9					
50	4	22	5	222	20				1	10					
51	4	22	5	222	20				1	11					
52	4	22	5	222	20				1	12					
53	4	22	5	222	20				1	13					
54	4	22	5	222	20				1	14					
55	4	22	5	222	20				1	15					
56	4	22	5	222	20				1	16					
57	4	22	5	222	20				1	17					
58	4	22	5	222	20				1	18					
59	4	22	5	222	20				1	19					
60	4	22	5	222	20				1	20					
.	.	.	.	.	.				.	.					
.	.	.	.	.	.				.	.					
.	.	.	.	.	.				.	.					
70	4	22	5	222	20				1	30					
27-APR-88		07:01 PM							CMD		CALC		CAPS		

The header abbreviations on the upper right of the data entry sheet stand for the following:

SX = Sex (See Table 1 for sex codes)
GI = Gonad Index

NOTE - Because the gonad index is very subjective and it is not used for data analysis, the gonad index was eliminated from the PWS herring sampling program in 1988.

LEN = Standard Length (to nearest mm)
WT = Weight (to nearest gm)
AGE = Age in years

NOTE - Numerical age codes are given in Table 1.

b. Entering Data:

Enter the weights, lengths, and sex data for the first 30 fish, then press the "ALT" key and the letter "C" key simultaneously. The (ALT)C macro will save the first 30 entries (e.g. AWL 1, Fish Nos. 1-30) of this new data file. After the file has been saved, the computer will display the word "READY" in the upper right corner of the monitors screen. At this point, press the [RETURN] key and the (ALT)C macro will kick in and ask:

HOW MANY FISH ON THIS CARD (NN) ?

Type "30" and then press the [RETURN] key. The (ALT)C macro will then enter the header information for the next 30 AWL data entries (e.g. AWL 2, Fish Nos. 1-30).

c. Using the (Alt)C Macro to Backup and Save the Data File:

After you enter the weights, lengths, and sex data for the second 30 AWL entries and each subsequent AWL form (e.g. AWL 3, Fish Nos. 1-30...), press the "Alt" key and the letter "C" key simultaneously again. This time and each time hereafter, the computer will display two options:

CANCEL REPLACE

In each case, you will want to press the letter "R" for replace. This will update the last file that you saved with the most recent set of 30 AWL data entries.

Again when the computer displays the word "READY" in the upper right corner of the monitor, press the [RETURN] key. Once again you will be asked:

HOW MANY FISH ON THIS CARD (NN) ?

Type "30" and then press the [RETURN] key. The (ALT)Cmacro will then enter the header information for the next 30 AWL data entries (e.g. AWL 3, Fish Nos. 1-30) etc.

d. Saving the Data File After the Sample is Completed:

When you complete your sample and all of the weight, length, and sex data has been entered, save your file by doing the following:

Press "/" (Displays the LOTUS 1-2-3 Command Menus)

Press the first letters of the following commands in the order that is indicated - FILE, SAVE

Enter the name of your new data file (8 Characters)

NOTE - At this point you must type in the name of your new data file (e.g. 88HTWBD1.WK1) or it will be saved as HAWLV288.WK1 which is the name of the last LOTUS 1-2-3 file that was retrieved. Refer to Section IV, Part B, No. 6a of the Methods for file naming conventions for data files.

Press [RETURN]

e. After you have saved your data file on the "original" data diskette, place it in the plastic sleeve in the black 3-ring binder entitled "1988 PWS Herring AWL Data and Summary Tables". Make sure that the diskette is labeled clearly (e.g. 1988 PWS Herring, Sac Roe Test P/S Catch, AWL Data and Summary Tables, Original).

f. Then make a "backup" copy of your data file on a second diskette by repeating the same procedures indicated in step (d) above. Again make sure that this backup diskette is clearly labeled and then insert it in the plastic sleeve below the original in the "1988 PWS Herring AWL Data and Summary Table" notebook.

7. Printing a Data File:

Next print out a hard copy of the data file you have created using the Compaq Deskpro computer and the Okidata printer in Drew's office. This hard copy can be used by the individual that is reading the scales, to record the ages of each fish.

NOTE - The scale reader also has the option of bringing the portable Compaq Plus computer into the scale reading room with him and entering the age data directly into the computer as he ages each scale.

a. To produce a hard copy of the data file, turn on the Compaq Deskpro computer and enter the LOTUS 1-2-3 subirectory by doing the following.

```
C:\>
C:\>CD\123
C:\123>123     (The computer will highlight "1-2-3")
```

b. Access the 1-2-3 spreadsheet program by pressing the [RETURN] key. A blank 1-2-3 spreadsheet will be displayed on the monitor.

c. The computer in Drew's office has been preset to operate off the A-Drive, therefore you will not have to change the FILE, DIRECTORY when you work on LOTUS files on this computer.

d. Insert the diskette which contains your LOTUS 1-2-3 data file (e.g. 88HTW801.WK1) into the A-Drive.

e. To retrieve this file for use, do the following:

Press the slash "/" key. (Displays the 1-2-3 Command Menus) Depress the first letter of the following commands in the order that is indicated - FILE, RETRIEVE (The computer will ask)

Name of file to retrieve: A:*.wk ?

and it will highlight the first file on the diskette. To call up this file, press the [RETURN] key. If there is more than one file on the diskette, use the "right cursor" key to move the highlighted cell to the file you wish to retrieve and then press the [RETURN] key.

The computer will display your data file as it was when you saved it last.

f. When you print out your data file, you should be sure to include the cell which contains the title of the data file (e.g. A38) and the cell which is two columns to the right of your last weight entry (e.g. P603) in the range of cells to be printed (e.g. A38..P603). To determine the range to be print do the following:

First determine the last cell in your data file by depressing the "end" key and the "right arrow" cursor key until you get to the weight column and then press the "end" key and the "down arrow" cursor key. This will reposition the cursor to the last weight entered in the lower right hand corner of the file. Now in order to produce a column for ages and to insure that the entire title of the file is printed out, move the cursor two spaces to the right with the "right arrow" cursor key. The position location of the cursor will be displayed in the upper left hand corner of the monitor screen. Record this cell location on a scrap of paper (e.g. P603). This will be the cell address you will indicate as the last cell in the range to be printed.

The first cell of your data file will be the cell where the title is located. To locate this cell do the following:

Press the "Home" key. (Will relocate the cursor to the upper left hand corner of your data file.)

Press the slash "/" key. (Displays the 1-2-3 Command Menus)

Depress the first letter of the following commands in the order that is indicated - WORKSHEET, TITLES, CLEAR

This series of commands will clear the title locks which have been preset to display the data entry headers as titles on the top line of the file.

Use the "up arrow" cursor key to move the highlighted cell up to cell A38 in your spreadsheet. This will be the first cell in the range that you will want to print. Therefore you have now determined that the range of cells to be printed for this sample data file are (A38..P603).

g. After you have determined the range of your file that will be printed, you are ready to do the following:

h. The data file can be printed out on the Okidata printer on 8 1/2 x 11 inch paper by doing the following:

Turn on the Okidata printer by depressing the switch on the lower right corner of the back of the printer. The red "SEL" and "POWER" lights on the left front panel are light when the unit is turned on and ready to go. Make sure the printer is loaded with 8 1/2 X 11 inch paper and that the ribbon shield on the printer head is even with the crease on the next clean sheet of paper.

Press the slash "/" key. (Displays the 1-2-3 Command Menus)

Press the first letter of the following commands in the order that they are indicated -

PRINT,
PRINTER,
RANGE (Type in the range that you have determine, e.g. A38..P603)

Press the [RETURN] key.

Press the first letter of the following commands in the order that they are indicated -

ALIGN
GO (The data file will be printed out by the printer)

8. Updating the AWL Data File With Age Data:

After the scales have been aged, the data file can be updated with the age data by doing the following:

a. Turn on the Compaq Deskpro computer and enter the LOTUS 1-2-3 subdirectory by doing the following.

```
C:\>
C:\>CD\123
C:\123>123     (The computer will highlight "1-2-3")
```

b. Access the 1-2-3 spreadsheet program by pressing the [RETURN] key. A blank 1-2-3 spreadsheet will be displayed on the monitor.

c. The computer in Drew's office has been preset to operate off the A-Drive, therefore you will not have to change the FILE, DIRECTORY when you work on LOTUS files on this computer.

d. Insert the diskette which contains your LOTUS 1-2-3 data file (e.g. 88HTW8D1.WK1) into the A-Drive.

e. To retrieve this file for use, do the following:

Press the slash "/" key. (Displays the 1-2-3 Command Menus)

Depress the first letter of the following commands in the order that is indicated - FILE, RETRIEVE
Name of file to retrieve: A:*.wk ?

The first file on the diskette will be highlighted. To call up this file, press the [RETURN] key. If there is more than one file on the diskette, use the "right arrow" cursor key to move the highlighted cell to the file you wish to retrieve and then press the [RETURN] key.

f. The computer will indicate "WAIT" in the upper right corner of the monitor while it is retrieving the file. When the retrieval is complete, the word "READY" will be indicated and you can cursor over to the AGE column and type in the age data.

g. After all of the ages have been entered ,resave the data file on the "original" data diskette and the "backup" data diskette.

h. Then print out a hard copy of the completed AWL data file (Table 2). When your going through the print commands, jot down the name of the file (e.g. 88HTWB01.WK1) and its range (e.g. A38..P603) on a piece of scrap paper. This information will be needed in the next step which is the production of a summary table.

NOTE - If you resave your data file after printing the hard copy, the print commands will also be saved with the file.

9. <u>Importing the Data File to Produce Summary Tables With the (Alt)P Macro</u>:

When your AWL data file is complete, you are now ready to import the data file back into the master macro file to produce a summary table.

a. Insert the diskette which contains the LOTUS 1-2-3 macro file named HAWLV288.WK1 into the A-Drive.

b. To retrieve this file for use, do the following:

Press the slash "/" key. (Displays the 1-2-3 Menus)

Depress the first letter of the following commands in

the order that is indicated - FILE, RETRIEVE

Name of file to retrieve: A:*.wk ?

The first file on the diskette, HAWLV288.WK1, will be highlighted . To call up this file, press the [RETURN] key.

The computer will display the upper left section of the HAWLV288.WK1 file on the monitor.

c. Remove the diskette which contains the HAWLV288.WK1 file from the A-Drive and insert the data diskette that contains the data file that you just completed (e.g. 88HTWBD1.WK1).

d. Use the "left arrow" and "down arrow" cursor keys to move the cursor to cell A38 in the spreadsheet.

e. To import the data file back into the macro file do the following:

Press the slash "/" key. (Displays the 1-2-3 Command Menus)
Depress the first letter of the following commands in the order that is indicated -

FILE,
COMBINE,
COPY,
NAMED/SPECIFIED RANGE

Enter range name or coordinates:

Type in the data files range (e.g. A38..P603)

Name of file to combine:

Type in the data files name (e.g. 88HTWBD1.WK1) and then press the [RETURN] key.

The word "WAIT" will flash in the upper right hand corner of the monitor screen while the data file is being imported into the macro. When the importing process is finished the data file will appear on the screen and the word "READY" will be indicated.
Use the cursor keys to check the file that was imported and make sure that it is complete.

f. To initiate the macro which generates the AWL summary table, press the "Alt" key and the letter "P" key simultaneously. This macro command will produce a

summary table of your data file which is located in the far upper right hand section of this spreadsheet (Range - AU1..BM25). The (ALT)P macro will then ask the following questions:

. ENTER BRIEF TABLE TITLE:

(e.g. PRINCE WILLIAM SOUND SAC ROE HERRING, WELLS BAY TEST PURSE SEINE SAMPLE, 10 APRIL 1988.)

NOTE - The following convention has been established for naming herring AWL summary tables for Prince William Sound:

Management Area (e.g. PRINCE WILLIAM SOUND)
Fishery (e.g. SAC ROE HERRING)
Sampling Location (e.g. WELLS BAY)
Sample Type (e.g. TEST)
Gear Type (e.g. PURSE SEINE)
Date (e.g. 10 APRIL 1988)

ENTER TABLE FILE NAME: (e.g. 88HTWBT1.WK1)

NOTE - The file naming conventions for summary tables is identical with those used for naming data files with one exception. Data files end with the letter "D" and then a series number and summary tables end with the letter "T" and then a series number.

g. After the computer indicates that it is once again "READY", do the following:

Press the "Home" key. (Will relocate the cursor to the upper left hand corner of your data file.)

Press the slash "/" key. (Displays the 1-2-3 Command Menus)

Depress the first letter of the following commands in The order that is indicated - WORKSHEET, TITLES, CLEAR
This series of commands will clear the title locks which have been preset to display the data entry headers as titles on the top line of the file.

Use the "Home" cursor key to relocate the cursor to the upper left hand corner of the spreadsheet.

Use the "End" key and the "right arrow" cursor key to move right in

the spreadsheet to the section where the summary table is located (Range AU1..BM25). Position the cursor at the upper left hand corner of the summary table (Cell AU1).

h. Printing Summary Table:

To print out a hard copy of the summary table, do the following:

Turn on the Okidata printer by depressing the switch on the lower right corner of the back of the printer. The red "SEL" and "POWER" lights on the left front panel are light when the unit is turned on and ready to go. Make sure the printer is loaded with 11 X 8 1/2 inch paper (Note - different paper size than data files) and that the ribbon shield on the printer head is 2 1/2 inches below the crease on the next clean sheet of paper. Also release the lock levers on the tractor unit and move them and the 11 x 8 1/2 inch paper to the far left setting.

Press the slash "/" key. (Displays the 1-2-3 Menus)

Press the first letter of the following commands in the order that they are indicated -

PRINT,
PRINTER,
RANGE (Range preset for AU1..BM25, need not be changed)

Press the [RETURN] key.

Press the first letter of the following commands in the order that they are indicated -

ALIGN
GO (The AWL summary table will be printed out by the printer)

An example of a typical AWL summary table that has been produced using the (ALT)P macro file option is given in Table 3.

V. Aging Scales

A. Committee Scale Aging Method:

During the 1988 Sac Roe Herring Season in Prince William Sound, scales from each herring sample were aged by a committee of two or three persons. Individuals which were involved in this committee aging

process were: Tom Schroeder (CF/Homer), Gene Sandone (CF/Anchorage), Henry Yuen (CF/Anchorage), Keith Schultz (CF/Bethel), Blaine McKnight (CF/Cordova), Diane Phipps (CF/Cordova), Sam Sharr (CF/Cordova), and Drew Crawford (CF/Cordova). Drew Crawford was the only member of this committee who was involved in the aging of all of the 1988 PWS sac roe herring scales, therefore he tried to maintain consistent aging practices between one group and the next. Scale images were projected on a microfiche screen and each individual aged the scales independently. If the scale readers arrived at a different conclusion regarding the age of a particular scale these differences were discussed and an age was determined by mutual consensus.

B. Scale Aging Conventions for PWS Herring:

Out of these discussions the following conventions were arrived at and used for aging all 1988 PWS sac roe herring scales.

1. On all herring which are caught and aged in late March through April, we assumed an annulus on the outer edge of each scale. Since each of these fish had just come through a winter, we felt that an annulus was or would be formed on the outer edge of the scale and therefore the outer edge was always counted as the most recent annulus (Figure 5).

   NOTE - On herring that are caught in the fall bait/food herring fishery in PWS, scale agers do not automatically count the outer edge as an annulus unless one is clearly visible.

2. On 99% of the scales that were aged, the focus was counted as the first annual growth ring.

3. Scales from younger age class fish (age 3 and age 4) which had a regenerated focus but exhibited strong growth on the outer margins were aged. Ages were assigned based on a comparison of the number of annuli that were visible and how the fishes length compared to other known fish of a similar age in the same sample. The primary assumption that was made in assigning an age to these scales was that there was a focus in the regenerated area in the center of the scale.

4. Regenerated scales from older age class fish (age 5 to age 12) were not aged.

5. Illegible scales were not aged.

6. Two irregular scale patterns emerged early in the 1988 PWS sac roe herring season (Figure 6).

   a. One of these irregular scale patterns was characterized by the formation of a small focus within an annual growth ring that was similar in size to the focus's of

other scales in the sample.

b. The second irregular pattern exhibited an annulus-like formation that was closer to the focus of the scale than the width of the distance between this annulus-like formation and the next annulus as measured outward from the center of the scale.

Since we were uncertain about the correct age of the fish with these irregular scale patterns, we decided to adopt two scale aging conventions that have been used by Tom Schroeder to age these types of herring scales in Cook Inlet.

Scale Aging Convention for (a) - For the infrequently observed scales that have a small focus within an annual ring that was similar in size to the majority of focus's in the sample, we decided to "discount the small focus" and begin our age count with the annual ring that was similar in size to the other focus's observed in that sample.

Scale Aging Convention for (b) - For the small number of scales that exhibit an annulus-like formation that was closer to the focus of the scale than the width of the distance between the annulus-like formation and the next annulus as measured outward from the center of the scale, we decided to call this annulus-like formation a "false check" and not count it as an annulus.

With regards to convention (a), we simply don't know what causes this type of scale pattern and whether this small focus should or should not be counted in the aging process. Therefore we identified the scale pattern and established this scale aging convention in order to be consistent about how this scale pattern is aged.

We can also not be certain whether convention (b) is right or wrong, however we have again identified a particular pattern and tried to be consistent about how we age it. However with regards to convention #2 we notice two things: (1) On the majority of scales that we age, the distance between the focus and the first annulus is usually large and the distance between the first annulus and the second is usually slightly smaller. Beyond the second annulus the distances between annuli were quite variable. (2) The lengths and weights of fish that exhibited a "false check" near its focus were consistent with the average lengths and weights in the age groups that they were assigned as a result of this aging convention.

Again we are not saying that these scale aging conventions are good or bad, we are simply attempting to document the procedures we used for aging herring scales in Prince William Sound in 1988. If anyone reading this document has additional information that

would aid in the aging of these two irregular scale patterns, please contact Drew Crawford at the ADF&G office in Cordova.

VI. Sample Size

A. At the start of the 1988 PWS Sac Roe Herring Season the standard sample size for an AWL sample was 600 herring. Midseason, Henry Yuen notice that out of a typical 600 fish sample that the number of scales that were tallied in the unaged (regenerated, illegible, missing) category was consistently at 5% or less of our total sample. He then suggested that as long as we were able to maintain the unaged scale category at that level, that we could reduce our sample size and still achieve the same degree of precision. Therefore, this idea was discussed with Project Leader, Sam Sharr and Biometrician, Linda Brannian and the sample size was reduced to 560 fish.

B. Post season analysis of 1988 PWS herring data by Linda Brannian, indicate that the optimal AWL sample size for Prince William Sound herring is 410 fish. This AWL sample size was increased to 450 herring to account for regenerated and illegible scales.

C. Collection of herring AWL samples on the fishing grounds:

1. Whenever possible collect samples from five different boats per area.

2. Collect 90 herring from each boat.

VII. Pooling Samples

Herring AWL samples from PWS are occasionally pooled.

A. One instance in which two small samples are pooled is when similar samples from the same location are received on consecutive days. If neither day's sample is large enough to constitute a full sample, these data are pooled to produce one sample.

B. Another instance in which samples are pooled is when multiple same-day samples are received from several locations that are in close proximity. In this case, work up a partial sample from all five boats in each area (e.g. 3 locations = 150 fish/area, 30 fish /boat; 2 locations = 225 fish/area, 45 fish/boat; etc.) and then test for age composition differences between areas using a chi-square test (Table 4). Only major age classes comprising 5% or more of the sample will be used in the chi-square analysis. If the age composition of the samples are not significantly different, the samples are pooled. If there are significant differences between the age compositions of the areas tested, then a full sample (450 fish/area, 90 fish/boat) is worked up for each area that is different.

Table 1. Gear codes, sex codes, and age codes for herring AWL sampling.

GEAR CODES:	1	Variable mesh gillnet, floating
	2	Variable mesh gillnet, sinking
	3	Set gillnet
	4	Drift gillnet
	5	Purse Seine
	6	Beach seine
	7	Otter trawl
	8	Hand picked
	9	Dip net
SEX CODES:	1	Male
	2	Female
	3	Juvenile
	4	Unknown
AGE CODES:	1-17	Actual fish age in years
	18	Regenerated
	19	Illegible
	20	Missing
	21	Wrong scale

Table 2. Sample printout of a Prince William Sound AWL data file.

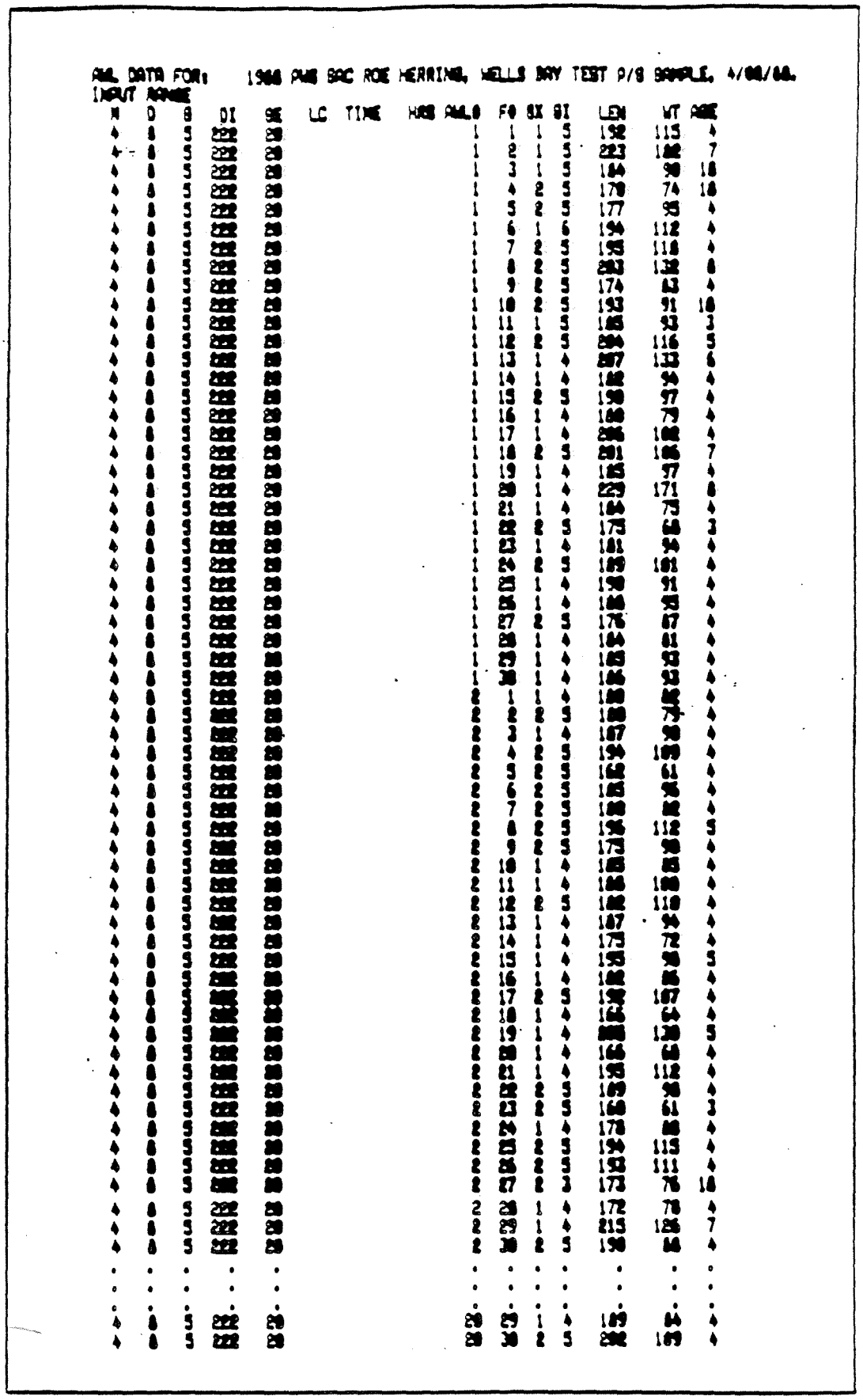


Table 3. Sample printout of a typical Prince William Sound herring AWL summary table.

SUMMARY TABLE : PRINCE WILLIAM SOUND BIG BOE HERRING, GALENA BAY TEST PURSE SEINE SAMPLE, 9 APRIL 1988.

	MALES						FEMALES						SEXES COMBINED					
			LENGTH		WEIGHT				LENGTH		WEIGHT				LENGTH		WEIGHT	
AGE	NUMBER	PERCENT	MEAN	STD	MEAN	STD	NUMBER	PERCENT	MEAN	STD	MEAN	STD	NUMBER	PERCENT	MEAN	STD	MEAN	STD
2	0	0.0	NA	NA	NA	NA	0	0.0	NA	NA	NA	NA	0	0.0	NA	NA	NA	NA
3	6	1.0	164	8	62	10	7	1.2	168	6	69	6	13	2.3	166	7	66	8
4	101	17.6	186	8	91	13	116	20.2	190	10	101	19	219	38.2	189	9	96	17
5	18	3.1	201	8	121	22	26	4.5	202	9	120	17	44	7.7	202	9	121	19
6	13	2.3	210	11	136	24	24	4.2	211	9	147	19	37	6.5	211	10	143	22
7	64	11.2	215	10	147	22	46	8.0	221	9	167	21	110	19.2	217	10	155	24
8	55	9.6	222	8	160	22	50	8.7	225	10	176	25	106	18.5	223	9	171	24
9	8	1.4	225	6	172	12	8	1.4	230	9	185	31	16	2.8	227	8	179	24
10	7	1.2	235	6	197	17	6	1.0	232	8	193	22	13	2.3	233	7	195	20
11	4	0.7	229	3	190	8	4	0.7	234	3	189	4	8	1.4	231	4	189	6
12	2	0.3	239	2	224	19	5	0.9	230	6	224	25	7	1.2	234	5	224	24
13	0	0.0	NA	NA	NA	NA	0	0.0	NA	NA	NA	NA	0	0.0	NA	NA	NA	NA
TOTAL	278	48.5	205	19	130	48	292	51.0	207	19	137	42	573	100.0	206	19	133	41
UNSEX	11	48.7	213	20	149	36	16	59.3	218	19	161	48	27	100.0	216	19	156	39

Table 4. Sample printout of a LOTUS 1-2-3 macrofile chi square test results for differences by age composition between two areas.

Chi-square analysis of the hypothesis that the frequencies of occurence of age are independent of area.

[redacted] Herring Age Data, 4/21/88.

Number of Herring

Area	Age 2	3	4	5	6	7	8	9	10	11	12	13	14	Total
[redacted]			[redacted]	27	13	26	15							[redacted]
[redacted]			[redacted]	17	8	15	8							[redacted]
Total			[redacted]	44	21	41	23							567

Expected numbers

[redacted]			217.1	21.8	10.4	20.3	11.4							
[redacted]			[redacted]	22.2	10.6	20.7	11.6							

Deviations

[redacted]			-17.1	5.2	2.6	5.7	3.6							
[redacted]			17.1	-5.2	-2.6	-5.7	-3.6							

Chi-square = 11.79

Chi-square = 9.488 4 d.f. and probability of a greater value = 0.05

Conclusions Frequencies of occurence of age are not independent of area.

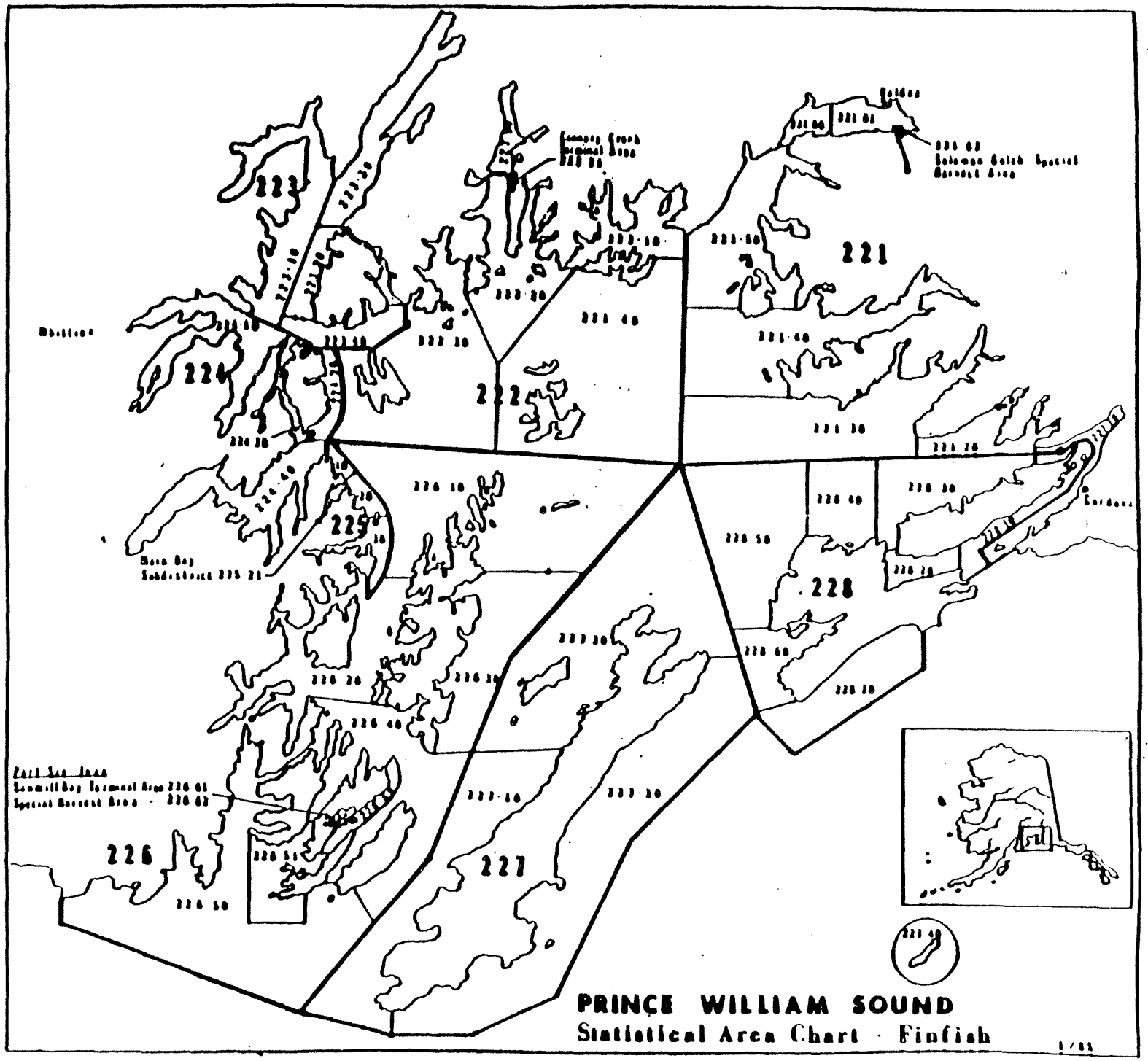


Figure 1. District and section location codes for Prince William Sound herring AWL sampling.

Standard Length: the straight line distance from the anterior most part of the fish, including the lower jaw with the mouth closed, to the end of the vertebra (hypural plate). The vertebra end is recognized by the folding of the skin on an unskinned fish when the tail is sharply bent.

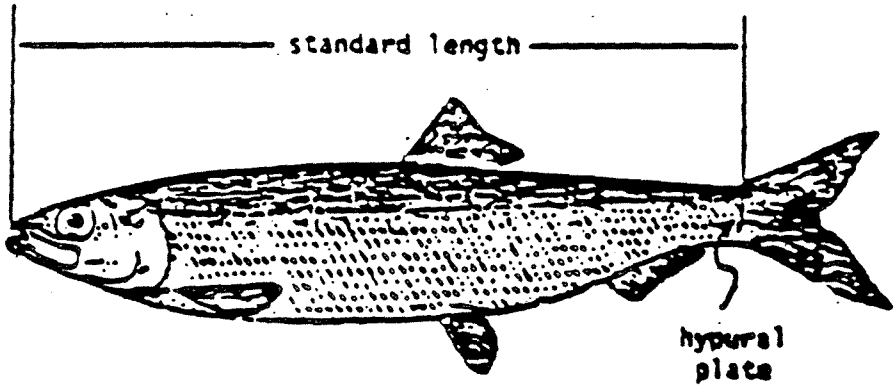
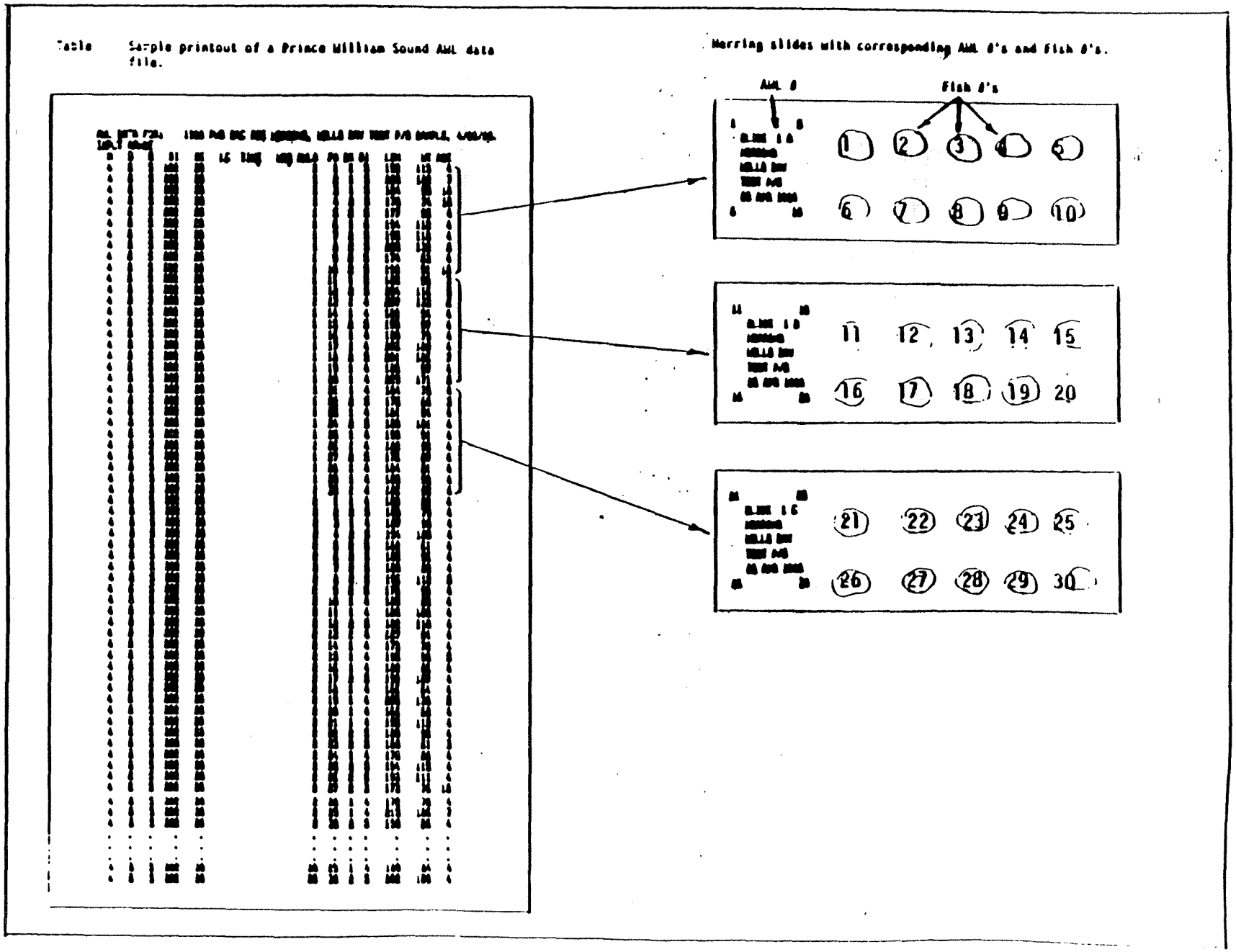
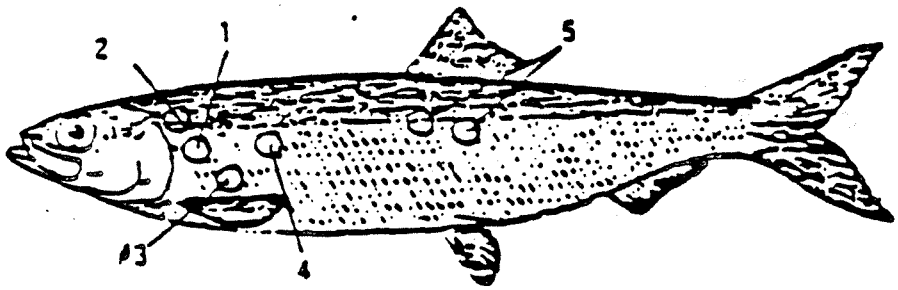


Figure 2. Standard length measurement for herring AWL sampling.

Figure 3. Proper scale placement on herring slides for aging and the relationship of the scale placement to its corresponding AWL data in the LOTUS 1-2-3 data file.





Prefered body areas from which to collect scales for aging are shown above. Scales should be collected from the left side of the herring if possible. If no scales are present in any of the above 5 areas on the left side, then check the right side (using the same preference sequence). If no scales are found on the right side, discard the herring and obtain another sample.

Figure 4. Preferred body areas from which to collect herring scales for aging.

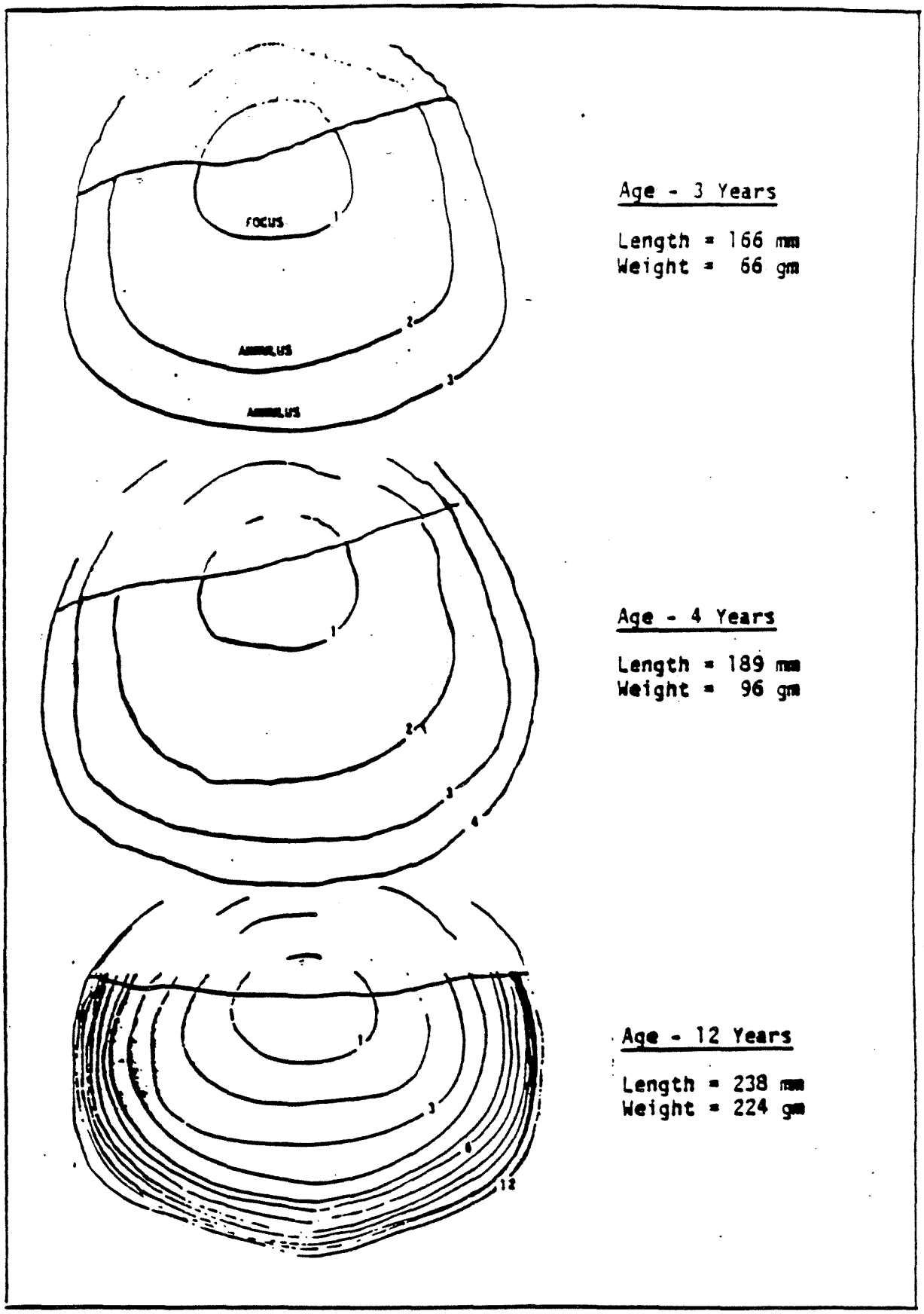


Figure 5. Sample Prince William Sound herring scale patterns and their corresponding age, length, and weight data.

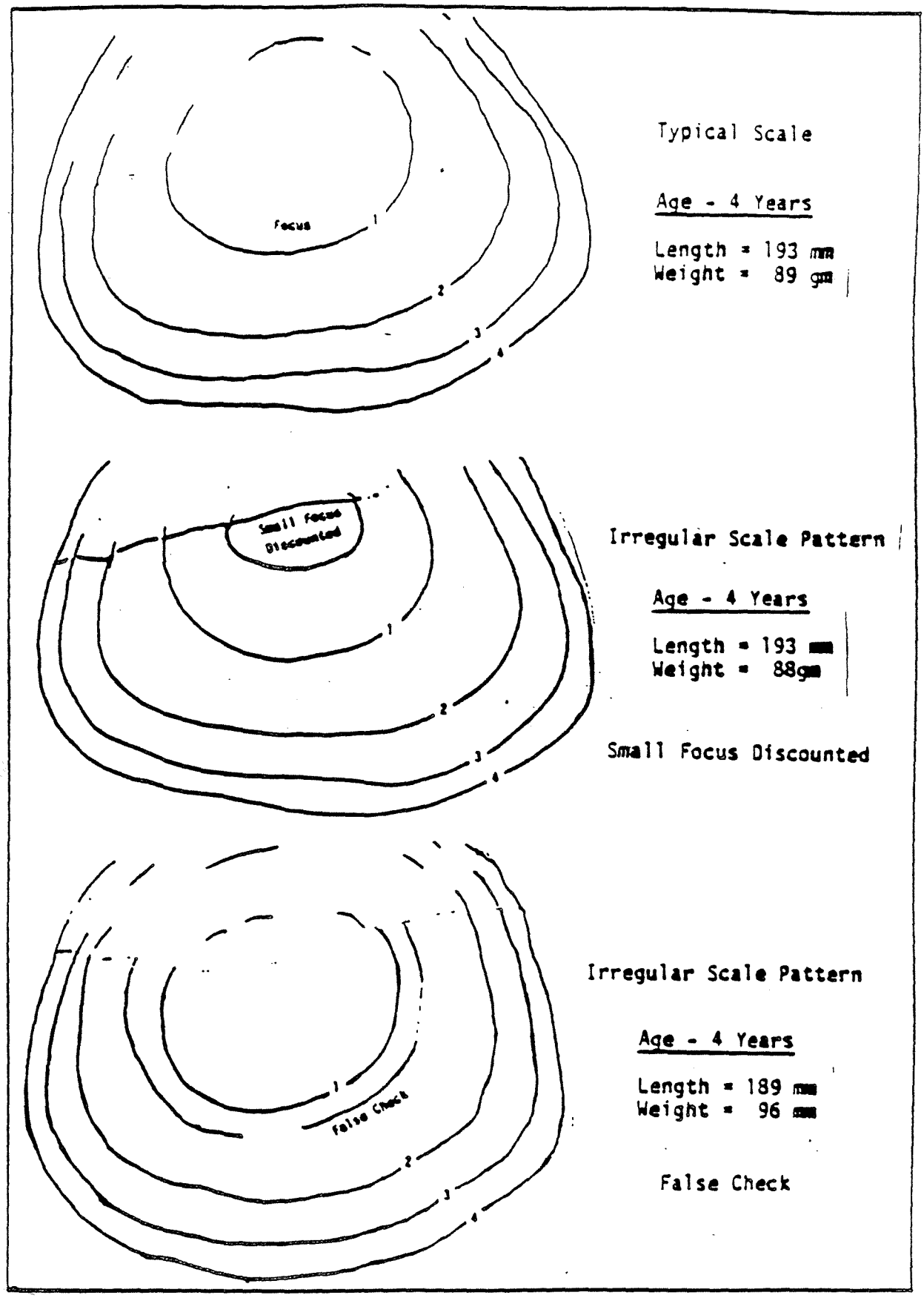


Figure 6. Comparison of a typical four year old Prince William Sound herring scale with scales from similar sized fish that exhibit irregular scale patterns.

APPENDIX E. Standard protocol for collecting herring eggs for hydrocarbon analysis (from J. Rice, NOAA/NMFS/Auke Bay Fisheries Lab).

Note: The first series of samples done under this protocol are adult herring obtained as a part of ADF&G's routine annual herring test fishing in Prince William Sound. This protocol can be used on other species also.

1 - Of primary importance is keeping all target tissues free of any contaminations which may interfere with credible analyses for hydrocarbons.

2 - Ancillary data (date of spawn, location, and depth of spawn) will be established during ADF&G's regular herring spawn deposition program. This data can then be accessed on their data base.

3 - SAMPLING: A small portion of roe on kelp will be taken by divers. Total of each sample will be approximately 10 grams, and each sample will be taken in triplicate and placed in four ounce hydrocarbon sample jars.

a. Clean jars with detergent and water; rinse or dip in some solvent (dichloromethane or methylene chloride - Spect grade, freon or 1,1,2-Trichloro-trifluoroethane - Spect grade) **NOTE: DICHLOROMETHANE is a contact hazard and ideally should be used under a hood; however, in the absence of a suitably equipped laboratory - use in a well ventilated area and wear gloves. If you get any on your skin, flood immediately with water.**
Rinse instruments after dissecting each fish.

b. To collect sample, take each sealed jar underwater and open, once under the surface just enough to let out the air, then reseal. Once at sampling site, open the jar and using a piece of surrounding kelp cradled in the dive glove, tear off approximately a ten gram section of roe (about a half of a four ounce jar) and place in jar and seal. Once brought to the surface, pour off excess sea water and label carefully.

d. Samples from the first depth treatment will be done in triplicate(see item 6 below).

e. Freeze samples immediately.

4 - LABELS. All labels should be written with waterproof pen and include the following:

- Date
- Species, tissue type
- Location (Site name, Coordinates if possible)
- Sample No. (your choice)
- Project/Agency
- Who Collected

Samples will be also be assigned a unique number when processed through the Auke Bay Laboratory for priority ranking and analysis.

5 - CHAIN OF CUSTODY. Transfer of samples from one person to another must be documented for legal and credibility purposes.
Ideally, you will be provided forms to use for each or each group of samples. You must fill out, sign and date this form when you release the samples to another person and that person must sign the sheet when receiving samples. The form travels with the samples.

6 - SAMPLE DESIGN. There will be 20 index transects set up for the herring egg mortality study. From each transect, 3 jars of herring eggs and 3 jars of mussels will be taken at the lowest site of mussel distribution in the intertidal (MLLM). This translates into 15 herring egg samples and 15 mussel samples from each of the four oil treatment levels (High, Medium, Low, and No Oil). This results in a total of 120 samples.

In addition, one transect from the High treatment series and one transect from the No Oil treatment series will be selected to examine hydrocarbon content difference between depth strata. 3 jars will be filled with herring eggs at each of the three following depth strata: 0 ft., -5 ft., -15 ft. This will result in an additional 18 samples.

CONFIDENTIAL

STATE/FEDERAL NATURAL RESOURCE DAMAGE ASSESSMENT
DETAILED STUDY PLAN

DRAFT

Project Title:	HYDROCARBON INJURY ASSESSMENT - KODIAK AND ALASKA PENINSULA HERRING
Study ID Number:	Fish/Shellfish Study Number 12
Lead Agency:	State of Alaska, ADF&G; Commercial Fisheries Division
Cooperating Agency(ies):	Federal: None State: None
Project Leader:	Kevin Brennan, Fishery Biologist
Assisting Personnel:	Dennis Gretch, Fishery Biologist Kim Rudge, Fishery Technician

Date Submitted: October 6, 1989

Principal Investigator:		10/9/89
Supervisor:		10/9/89
OSIAR Senior Biometrician:		
OSIAR Program Manager:		10-13-89
OSIAR Director:		10-17-89

INTRODUCTION

The oil spill from the March 24, 1989 grounding of the M/T Exxon Valdez in Prince William Sound reached Kodiak and the Alaska Peninsula during the annual spawning migration of herring into shallow nearshore waters. Because herring spawn on intertidal and subtidal vegetation, mature spawning herring and their eggs and larvae are all particularly susceptible to injury. It is hypothesized that the oil spill will adversely impact adult fish through direct mortality, food shortage, slowed growth, and a possible reduction in fecundity. Possible effects on herring eggs and larvae include reduced egg survival, reduced hatching success and reduced viability of fry. The goal of this project is to determine whether the Exxon Valdez oil spill will have a measurable impact on Pacific herring (*Clupea harengus pallasi*) stocks of the Kodiak archipelago and the Alaska Peninsula.

Herring populations of the Kodiak archipelago and the Alaska Peninsula are composed of a large number of relatively small, discrete stocks. Because of the numerous, widely-separated areas in which herring spawn there are no comprehensive programs, such as spawn deposition surveys or aerial surveys, to determine the absolute abundance of herring in these areas. Because of the expense involved in surveying the large area, injury to herring in the area from the Exxon Valdez oil spill cannot be feasibly determined using abundance measures.

This study, in part, seeks to assess injury to Kodiak and Alaska Peninsula herring by applying the results of comprehensive damage assessment studies in Prince William Sound to similarly affected areas in the Kodiak archipelago and the Alaska Peninsula. A second part of the study will examine the growth rates of herring in affected and unaffected areas as compared with historical and future growth rates.

OBJECTIVES

1. Document the occurrence of herring stocks and spawn in oiled and non-oiled areas of the Kodiak Archipelago and the Alaska Peninsula.

2. Estimate the injury to herring eggs and larvae by directly applying results from Prince William Sound injury assessment studies.

3. Test the hypothesis that incremental growth by age is independent of oil impacts.

4. Identify potential alternative methods and strategies for restoration of fishery stocks, and/or habitat where injury is identified.

METHODS/DATA ANALYSIS

Coastal habitat injury assessment studies (Coastal Habitat Study #1), in conjunction with air/water resources injury assessment studies (Air/Water Studies #1-4), will determine the extent of potential injury, or degree of impact, by habitat type and level of oiling (unoiled, light, or moderate/heavily oiled). Studies in Prince William Sound seek to determine the effect of the oil impacts on adult herring growth and fecundity, on egg deposition, and on egg and larvae survival and development, through field and laboratory studies (Fish/Shellfish Study #11). If a quantifiable damage can be assessed from the Prince William Sound studies then these results will be directly applied to the spawning stocks in similarly impacted areas of the Kodiak/Alaska Peninsula area.

ADF&G Fishery Biologists will conduct fixed wing aerial surveys daily, as weather conditions and availability of charter aircraft permit. The survey area will comprise all of the Kodiak and Alaska Peninsula areas, but special emphasis shall be placed on those locations where herring spawn has been reported or stocks are reaching sexual maturity, as indicated by ADF&G field personnel, commercial fishermen or spotters, or where the "normal" timing of stocks would indicate. Aerial survey data will be recorded on the form listed in Appendix A. The location, number, relative size (small, medium, large, etc.), and an estimate of total biomass observed will be noted for all schools of herring and forage fish. Location, duration, and size of spawns will also be recorded. Similar information will be collected by ADF&G herring field crews, recorded in Rite-in-the-Rain notebooks, and reported to the Kodiak ADF&G office during daily radio contacts. Additionally, observations from reliable fishermen, spotters, or oil assessment crews will be evaluated and included where possible. Maps showing areas where pre-spawning herring have historically aggregated and where herring have consistently spawned will be prepared. Maps showing the distribution of milt along the shoreline will also be prepared. The impact of oil to various areas will be evaluated using the same criteria developed in Prince William Sound.

Since 1979, ADF&G has conducted intensive age-weight-length-sex (AWL) sampling of Kodiak/Alaska Peninsula herring stocks during the sac roe fishery. Approximately 4000-9000 fish are sampled each year. Burkey and Reid (1988a) report detailed results of the (AWL) sampling from 1981-87, and Burkey and Ried (1988b) summarize the available fishery harvest and age composition data from 1964-87. Hence, a large "pre-spill database" exists against which future growth rates can be compared.

During the upcoming commercial fishing seasons an attempt will be made to collect at least two 60 fish samples at different times in each of the 51 statistical areas (Figure 1). Detailed methodology used for collecting AWL samples is described in Appendix B, "Westward Region Herring Procedures for Collecting Data from Test and Commercial Fishing Catches". Samples will be collected from the commercial seine catch by ADF&G field personnel during the course of their usual duties, or, in cases where that is not possible, by rigging ADF&G's R/V Coho to operate a small midwater sampling trawl. All sampled fish will be measured for standard length (tip of snout to hypural plate) to the nearest millimeter, weighed to the nearest gram, and aged by analysis of scale annuli using a binocular microscope at 12X magnification. General linear model extensions of analysis of variance models will be used to test for year, area, and oil spill effects on herring growth.

LITERATURE CITED

Burkey, C. Jr., and J. Reid. 1988a. Age, sex, and size composition of Pacific herring from the Kodiak area, Alaska, 1981-87. Regional Information Report 4K88-34, Alaska Department of Fish and Game, Kodiak, Alaska.

Burkey, C. Jr., and J. Ried. 1988b. Statistics of the commercial fishery for Pacific herring from the Kodiak Area, Alaska. Technical Fishery Report 88-11, Alaska Department of Fish and Game, Juneau, Alaska.

SCHEDULES AND REPORTS

DATE(S)	ACTIVITY
April-June 1989	Aerial surveying, data collection
June-December 1989	Data analysis, map preparation
December 21, 1989	Final report
January-February 1990	Continuing investigation

PROJECT BUDGET

LINE ITEM	CATAGORY	BUDGET
100	Personnel	$ 15,000
200	Travel	
300	Contractual	$ 12,000
400	Supplies	$ 8,500
500	Equipment	
TOTAL		$ 45,500

FUNDED PERSONNEL

CLASS	PCN	NAME	MM	OT	SEA	HAZ	COST
FB II	7017	Kevin Brennan	2.0	0	0	0	$ 8,804.81
FB I	1844	Dennis Gretch	0.5	0	0	0	$ 2,074.35
FT III	1843	Kim Rudge	1.0	0	0	0	$ 3,359.80
TOTAL			3.5	0	0	0	$14,238.96

CONTRACTUAL SERVICES

ITEM	PROPOSED EXPENDITURE
73864 - Aircraft Charter	$ 10,000
(for aerial surveys and collection of samples from field crews)	
73780 - Repair/Maintenance Labor	$ 2,000
(for repair of winch and hydraulics of R/V Coho's trawl gear)	
TOTAL	$ 12,000

COMMODITIES

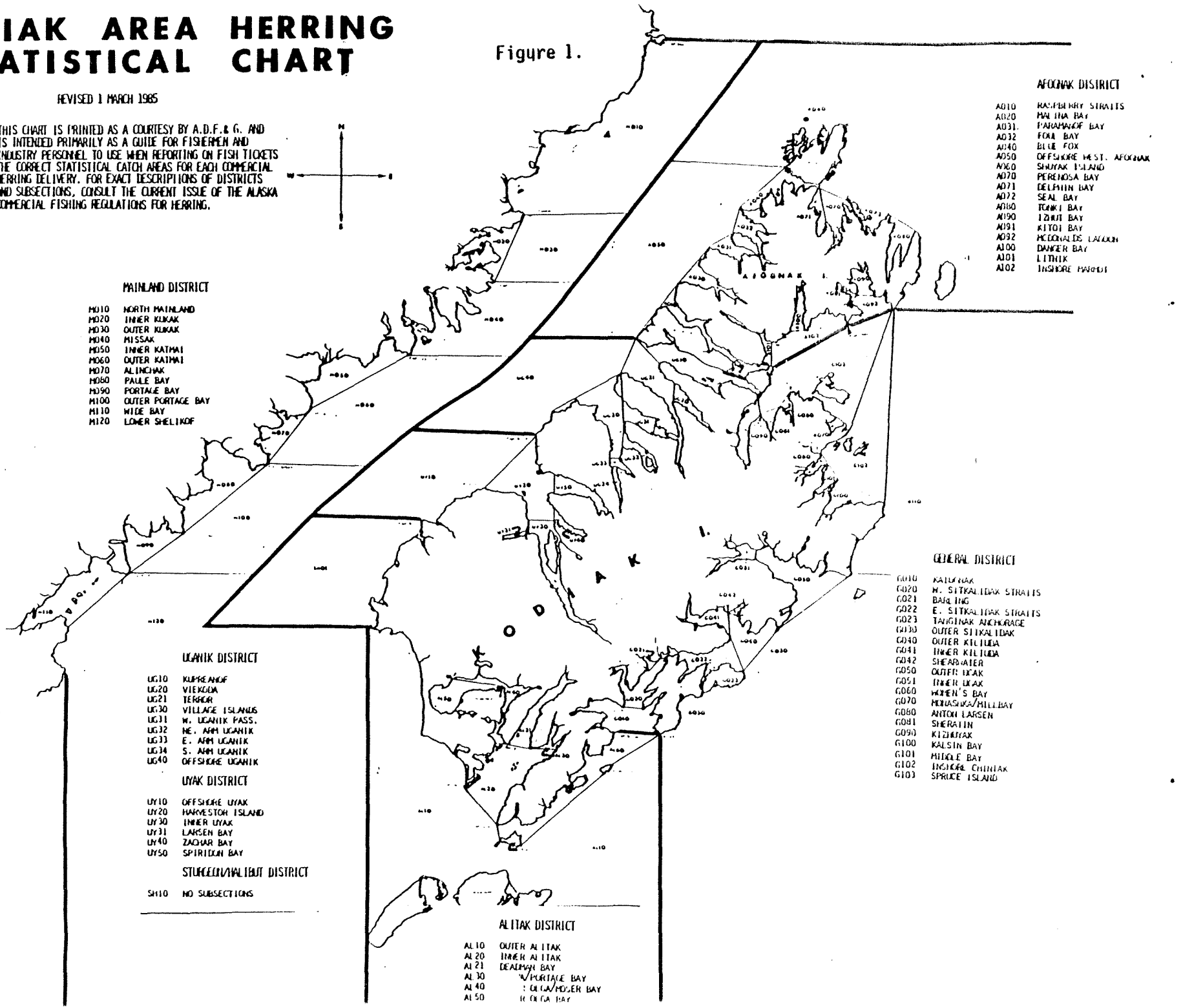
ITEM	PROPOSED EXPENDITURE
74229 - Office Supplies	$ 500
74520 - Scientific Supplies	$ 500
74616 - Marine Supplies	$ 7,500
(for new hydraulic motor, cables, etc., for R/V Coho's trawl)	
TOTAL	$ 8,500

KODIAK AREA HERRING STATISTICAL CHART

Figure 1.

REVISED 1 MARCH 1985

THIS CHART IS PRINTED AS A COURTESY BY A.D.F.& G. AND IS INTENDED PRIMARILY AS A GUIDE FOR FISHERMEN AND INDUSTRY PERSONNEL TO USE WHEN REPORTING ON FISH TICKETS THE CORRECT STATISTICAL CATCH AREAS FOR EACH COMMERCIAL HERRING DELIVERY. FOR EXACT DESCRIPTIONS OF DISTRICTS AND SUBSECTIONS, CONSULT THE CURRENT ISSUE OF THE ALASKA COMMERCIAL FISHING REGULATIONS FOR HERRING.



AFOGNAK DISTRICT

A010	RASPBERRY STRAITS
A020	MALINA BAY
A031	PARAMANOF BAY
A032	FOUL BAY
A040	BLUE FOX
A050	OFFSHORE WEST. AFOGNAK
A060	SHUYAK ISLAND
A070	PERENOSA BAY
A071	DELPHIN BAY
A072	SEAL BAY
A080	TONKI BAY
A090	IZHUT BAY
A091	KITOI BAY
A092	MCDONALDS LAGOON
A100	DANGER BAY
A101	LITNIK
A102	INSHORE MARMOT

MAINLAND DISTRICT

M010	NORTH MAINLAND
M020	INNER KUKAK
M030	OUTER KUKAK
M040	MISSAK
M050	INNER KATMAI
M060	OUTER KATMAI
M070	ALINCHAK
M080	PAULE BAY
M090	PORTAGE BAY
M100	OUTER PORTAGE BAY
M110	WIDE BAY
M120	LOWER SHELIKOF

GENERAL DISTRICT

G010	KATMAI
G020	W. SITKALIDAK STRAITS
G021	BARLING
G022	E. SITKALIDAK STRAITS
G023	TANGINAK ANCHORAGE
G030	OUTER SITKALIDAK
G040	OUTER KILIUDA
G041	INNER KILIUDA
G042	SHEARWATER
G050	OUTER UGAK
G051	INNER UGAK
G060	HOKEN'S BAY
G070	MONASHKA/HILL BAY
G080	ANTON LARSEN
G081	SHERATIN
G090	KIZHUYAK
G100	KALSIN BAY
G101	MIDDLE BAY
G102	INSHORE CHINIAK
G103	SPRUCE ISLAND

UGANIK DISTRICT

UG10	KUPREANOF
UG20	VIEKODA
UG21	TERROR
UG30	VILLAGE ISLANDS
UG31	W. UGANIK PASS.
UG32	NE. ARM UGANIK
UG33	E. ARM UGANIK
UG34	S. ARM UGANIK
UG40	OFFSHORE UGANIK

UYAK DISTRICT

UY10	OFFSHORE UYAK
UY20	HARVESTOH ISLAND
UY30	INNER UYAK
UY31	LARSEN BAY
UY40	ZACHAR BAY
UY50	SPIRIDON BAY

STURGEON/HALIBUT DISTRICT

SH10	NO SUBSECTIONS

ALITAK DISTRICT

AL10	OUTER ALITAK
AL20	INNER ALITAK
AL21	DEADMAN BAY
AL30	W/PORTAGE BAY
AL40	E OLGA/MOSER BAY
AL50	W OLGA BAY

APPENDICES

Appendix A. Kodiak Area herring aerial survey form.

FORAGE FISH AERIAL SURVEY REPORTS

OBSERVER:_____ DATE_____ PILOT:_____

TIME:_____ PLANE:_____

TIDE:_____ WEATHER:_____

Actual Location	Mgmt. Unit	HERRING SCHOOLS			VISIBILITY				MILT OBSERVED		GEAR OBSERVED				FORAGE FISH		MISCELLANEOUS COMMENTS
		No.	Tons/School	Total Tons	P	F	G	E	Length	Width	01	34	00	99	No. Schls	Tons	

Appendix B

Westward Region Herring Procedures for Collecting Data from Test and Commercial Fishing Catches

INTRODUCTION

Herring from the commercial catch and test fishing are sampled for sex, size and age annually by field crews in coastal waters of the State from Ketchikan to Kotzebue to form a data base essential to the management of the State's herring resources. This information is drawn upon by management and research biologists for monitoring and regulating harvest levels, determining run timing, entry patterns and distribution of herring arriving on the spawning grounds, monitoring sexual maturity and age composition of herring spawning populations, developing methods to forecast herring abundance and determine optimum spawning goals, and to gain a better understanding of the biology of each stock. The usefulness of this AWL data depends on the manner and accuracy in which the samples were taken.

This manual was prepared as a guide for collecting information on age, sex, maturity, length, weight and fecundity of herring and recording the appropriate data on A-W-L mark-sense forms. Also included are methods used for collecting similar information for capelin and other fishes encountered during herring sampling. The sampling and recording procedures are listed in a logical order of activity and should be followed in sequence to develop accurate sampling techniques.

Responsibility for accuracy lies first with the primary data collector(s). Above all, KEEP THE MARK-SENSE FORMS FLAT, DRY AND CLEAN. Fish gurry and water curling will cause data to be incorrectly read. Project supervisors will return sloppy or incomplete data to individual collectors. Each form shall be marked with the data recorder's and interpreter's initials.

When using the reverse side of the mark-sense form to record data, the sheet code must be transferred on to the extreme left hand column of the back page. To transfer the code, fold the form over (without creasing) so that both code columns are visible and mark the corresponding number blocks onto the back grid.

Please read all the instructions carefully so that information is collected and recorded accurately and properly. Process all fishes within 48 hours of capture. Use a number 1 (or 2) lead pencil to record data and write labels. Please print legibly.

If you have comments or suggestions for improving sampling methods or this manual, please contact Larry Malloy or Len Schwarz.

EQUIPMENT LIST

1. Plain glass microscope slides (25 x 75 x 1mm)
2. Forceps
3. Scissors
4. Scalpel

5. Lead pencils (number 1 or 2)

6. Small beakers (at least two)

8. Wide mouth sample jars containing Gilsons Fluid (fecundity samples)

8. Ziplock plastic bags

9. Measuring board (calibrated in mm)

10. Dial-a-Gram or Lume-O-Gram scale

11. Small vials with caps (otolith sampling)

12. Labels

13. Paper towels

14. Mucilage glue

15. Water

16. Ethanol or glycerol (otolith sampling)

17. A-W-L Mark-Sense data forms

18. Formalin

19. Eyedropper (otolith sampling)

20. Stick-on white labels 5/8 inches by 7/8 inches

METHODS

Herring

I. AWL Labeling

A. Before any fish are handled, label the waterproof A-W-L mark-sense form(s) with a soft No. 1 or (2) pencil referring to (Figures 1a, 1b, and 1c).

1. Leave the mark sense spaces blank if you are uncertain of a factor i.e. district number. These will be filled in in town.

2. Enter the following information in the blank space between the LOCATION heading and the WEIGHT and AGE heading.
   a. Samplers name
   b. Sample number: each sample should start with the crew leader's initials and then be numbered sequentially by sample.
   c. Location: the exact location where the sample was CAUGHT is very important. Note the distance from

the nearest headland (i.e. 2 miles N.E. Rocky Point). In the Port Moller and Canoe Bay fisheries refer to chart provided (Figure 2 and 2A) and record the nearest headland and subsection number.

d. Date: record the date the fish were CAUGHT on

e. Source: record how the fish were taken. i.e. commercial seine sample, variable mesh gillnet, seine test set, etc.

f. Vessel: record the name of the vessel the sample was taken from.

g. Collector: record the name of the person who collected the sample from the vessel or cannery.

3. Additional information should be entered on the back of the AWL form under comments. Such comments could include estimated size of delivery, roe percentage, or other background information. If a sample contains more than 30 fish it will require more than one AWL form. In the top right hand corner write in which page of the sample it is (i.e. page 1 of 3; in other words this is the first page of a sample that contains 3 pages).

II. Filling out AWL Forms

A. When entering data, blacken the entire block for its full length and width (▮). Be sure and mark all the way from the top to the bottom.

1. A single light pencil mark in the block is not acceptable (▯).

2. If a mistake is made, erase the mistake as completely as possible without rubbing away the paper and be sure to thoroughly blacken the correct mark to prevent a data entry error when the forms are machine processed.

B. Refer to the area code for a listing of designated A-W-L mark-sense codes (Figure 1b and 1c).

III. Labeling slides

A. Determine the number of microscope slides needed for mounting herring scales.

1. Scales will be mounted on plain slides with labels on one end.

2. Each slide will contain a maximum of 10 scales. One scale from each of 10 herring.

B. Label each slide with 5 lines of information (Figure 1a).

1. General Area. Write in either North or South Peninsula, Kodiak, Chignik, etc.

2. Catch location. Enter the exact location the fish were taken in. Do not record where the tender is but where the fish were harvested.
3. Catch date. Enter the date that the fish were harvested on.
4. Sample number. Enter the sample number. This should correspond with the sample number on the AWL form you are using.
5. Scale Number. Scale numbers should start with 1 and continue in multiples of 10. (i.e. 1-10, 11-20, 21-30, 31-40, 41-50, 51-60). If a sample contains 90 samples there will be 3 AWL forms labeled page 1 of 3, page 2 of 3, and page 3 of 3. Page 1 will have length and weight data corresponding to scales 1-30, Page 2 31-60, and Page 3 61-90.

C. Each sample and day should begin with a new AWL form.

IV. Measurements and Observations

A. IMPORTANT: After each herring is measured, weighed, sexed, etc., it should be placed aside in the same order it was sampled so that the scale taken will correspond to the correct herring specimen number (Figure 1a).

B. Standard length must be measured for every herring sampled unless the specimen has been too badly mangled (Figure 3).

1. Place each herring on the measuring board so that its anterior extremity is against the stop at the 0 mm line of the ruler (make sure the herring's mouth is closed).

2. Locate the area where the audal (tail) fin rays meet the hypural plate by sharply bending the tail and noting the location of the crease.

3. Record the measurement (to the nearest mm) from the anterior most extremity of the herring (the tip of the lower jaw) to the middle of the crease formed when the tail is bent.

4. Length is recorded on the A-W-L form by marking the appropriate column blocks.

C. Sex must be determined for every herring sampled, and appropriate column 1-4 marked under the SEX heading.

1. If herring are not ripe and running eggs or milt, they must be dissected to visually inspect the gonads.

2. If herring are immature (virgin), it is not possible to determine the sex without use of a microscope. Simply record these specimens as juveniles on the A-W-L form coded as 3.

3. If specimens cannot be sexed for some other reason(i.e. specimen badly mutilated, etc.) record as unknown on the A-W-L form coded as 4.

D. Sexual maturity index must be determined for every specimen whenever possible. Use the guide (Figure 1c to identify the appropriate Gonad Maturity number and mark the corresponding GONAD INDEX column block.

V. Scale Sampling

A. Only one readable scale will be taken from each herring with a maximum of 10 scales placed on each slide (Figure 1a).

B. Remove each scale from one of the preferred body areas (Figure 3) on the left side of the fish (right side used as alternate site if necessary) using forceps. Body area locations are numbered in order of preference (location 1 is most preferred; location 3 is least preferred). If stock separation is intended find a scale from the blackened area only (Figure 3) and mark P in the margin next to that fish. You will be instructed if this is necessary.

C. Dip each scale in clean water, rub between thumb and forefinger to remove dirt and slime, examine (hold up to a light) for regeneration (regenerated scales appear blurred in the center), DISCARD IF REGENERATED REPEATING PROCEDURE UNTIL A SUITABLE SCALE IS LOCATED.

D. To mount a scale on the glass slide, dip the scale into the mucilage glue solution, (1 part mucilage glue: 10 parts water), shake off excess solution, and place the scale onto the slide making sure the unsculptured (concave) side of the scale is facing down and the anterior margin (portion embedded in the integument of the fish) is facing towards the bottom of the slide. The ridges on the sculptured side of the scale can be felt with a fingernail or forceps. Make certain that scales are placed on the slides in the positions corresponding to the correct specimen number on the A-W-L form (Figure 1a).

E. Press each scale firmly against the slide with a paper towel after mounting to remove excess glue from underneath the scale. Press firmly and blot excess glue with the towel. (Too much pressure, however can break your slide.)

F. Store completed scale mounts in slide boxes to avoid loss or breakage.

G. Mark the age of each specimen in the appropriate blocks on the A-W-L forms after the scales have been aged. A regenerated scale is marked as 18, illegible as 19 and missing as 20 in the age column.

H. Completed A-W-L mark-sense forms should be stored in an appropriate notebook, file, etc.

VI. Otolith Sampling (see following Capelin sections I and III).

A. Do Not Collect otoliths unless directed to in the project operational plan.

CAPELIN

I. Preparations Prior to Data Collection (Do Not Collect otoliths unless directed to in your project operational plan)

A. Mix a solution of 1 part water to 1 part ethanol or use 100% glycerol.

B. Determine the number of vials needed for collecting otoliths.

1. Each vial will hold one pair of otoliths.
2. Thirty vials are required for every complete A-W-L form.

C. Place a label in each vial with the following information.

1. Sample number. This should correspond with the sample number on the AWL form being used.
2. Otolith Number. This should correspond with the A-W-L number which contains the appropriate length and sex for that otolith (i.e. 1-30 on the first page of the sample, 31-60 on the second page of the sample, etc.)
3. Species. Either capelin or herring.
4. Date. Put the date the fish were captured on.
5. Location. Put the area of capture
6. Sampler's name.

D. Make certain that capelin samples from test fishing catches have been correctly subsampled and contain equal numbers of males and females (see Test Fishing-Variable Mesh Gillnets, Operational Plan).

II. Measurements and Observations

A. IMPORTANT: After each capelin is measured, weighed, sexed, etc. it should be placed side in the same order it was sampled so that the otoliths taken will correspond to the correct capelin specimen number (Figure 4). All capelin data will be recorded on A-W-L mark-sense forms similar to herring data.

B. Fork length must be measured for every capelin in a sample, unless the specimen has been too badly mangled (Figure 5).

1. Place each capelin on the measuring board so that it's anterior extremity is against the stop at the 0 mm line of the ruler (make sure the capelin's mouth is closed).

C. Weight to the nearest g must be taken for every capelin in the sample, unless a portion of the body is missing.

D. Sex must be determined from every capelin in the sample, whenever possible.

1. Sexually mature males can be distinguished by the prominent raised "hairy" bands of scales along their sides (above the lateral line).

2. If capelin are not ripe and running eggs and sperm (and if males cannot be sexed externally), they must be dissected so that gonads can be visually inspected.

3. If specimens cannot be sexed, indicate this on the A-W-L form. Do not record these specimens as juveniles unless you have good reason to believe they are immature.

E. Sexual maturity index must be determined for every specimen whenever possible. Use the appropriate code (Figure 1C). Although the code guide was developed specifically for herring, it is general enough to use for most fishes, if specific gonad measurements are ignored.

III. Otolith Sampling

A. One pair of otoliths will be taken from each capelin and placed in an individual vial.

B. To find the otoliths make a shallow, horizontal cut beginning just behind the head and extending to the snout. This will expose the brain cavity (Figure 4).

C. The otoliths will be found on either side of the skull behind the eyes. (There are three pairs of otoliths located within the chambers of the inner ears. The largest pair, sagitta, are the easiest to find and remove.)

D. Remove the otoliths with forceps and clean each specimen between your fingers.

E. Place the pair in an appropriately labeled vial, fill with the ethanol solution or glycerol and cap tightly.

F. If an otolith breaks during the extraction process, place all pieces in the vial.

Other Fishes

Other fishes, such as yellowfin sole, may also need to be sampled in some areas. Process these specimens using the methods outlined for capelin (see above). However, use fork length on species with forked tails and total length on species that do not have forked tails. Indicate on A-W-L form under "remarks" when total length is used.

Figure 1A.

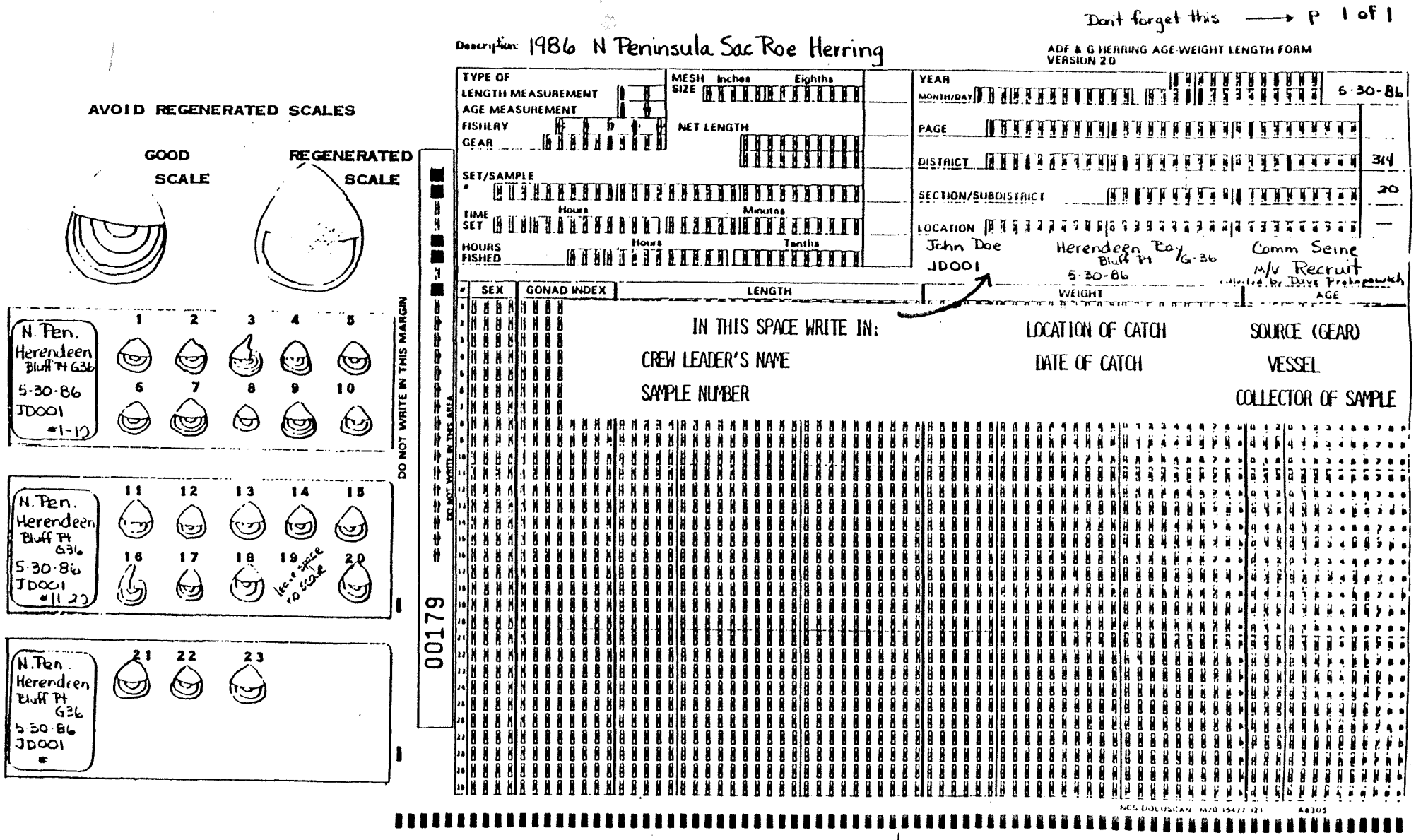
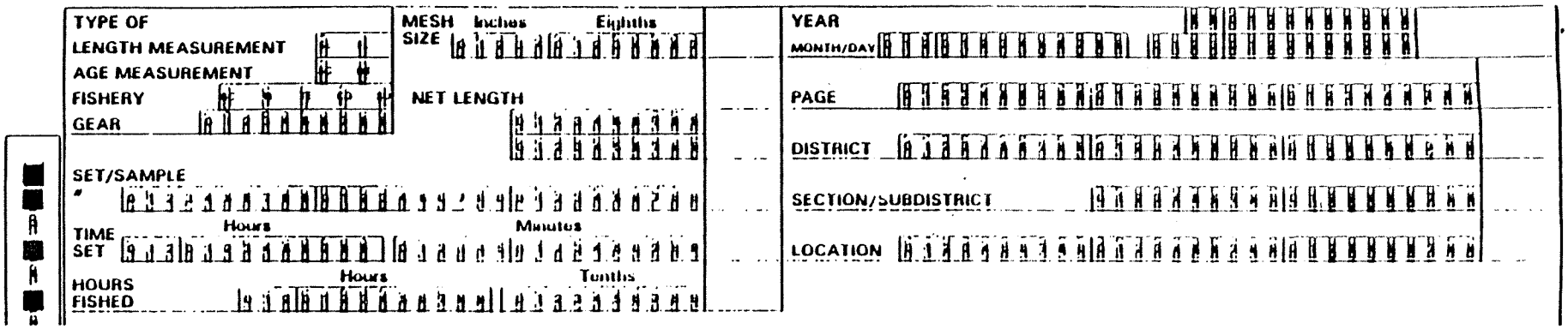

Don't forget this → P 1 of 1
Description: 1986 N Peninsula Sac Roe Herring
ADF & G HERRING AGE-WEIGHT-LENGTH FORM VERSION 2.0
AVOID REGENERATED SCALES
GOOD SCALE
REGENERATED SCALE
TYPE OF LENGTH MEASUREMENT
AGE MEASUREMENT
FISHERY
GEAR
MESH SIZE Inches Eighths
NET LENGTH
SET/SAMPLE #
TIME SET Hours Minutes
HOURS FISHED Hours Tenths
YEAR
MONTH/DAY 6-30-86
PAGE
DISTRICT 314
SECTION/SUBDISTRICT 20
LOCATION
John Doe
JD001
Herendeen Bay / Bluff Pt / G-36
6-30-86
Comm Seine
m/v Recruit
Collected by Dave Prokopowich
SEX
GONAD INDEX
LENGTH
WEIGHT
AGE
IN THIS SPACE WRITE IN:
CREW LEADER'S NAME
SAMPLE NUMBER
LOCATION OF CATCH
DATE OF CATCH
SOURCE (GEAR)
VESSEL
COLLECTOR OF SAMPLE
DO NOT WRITE IN THIS MARGIN
00179
DO NOT WRITE IN THIS AREA
N. Pen. Herendeen Bluff Pt G36 5-30-86 JD001 #1-10
N. Pen. Herendeen Bluff Pt G36 5-30-86 JD001 #11-20
Leave space to scale
N. Pen. Herendeen Bluff Pt G36 5-30-86 JD001 #
HERRING 1
2
3
4
11
12
21
22
23
HERRING SHOULD BE POSITIONED IN ROWS SO AS TO MATCH ENTRIES ON AWL FORMS
LEAVE A SPACE ON THE SLIDE FOR HERRING YOU CAN'T GET A SCALE FROM

Figure 1B.

P ___ of ____.

Description:

ADF & G HERRING AGE-WEIGHT-LENGTH FORM
VERSION 2.0



DESCRIPTION - Accurately state where sample was taken.

YEAR-MONTH-DAY - Date on which samples were collected. Remember - only one date per AWL.

LENGTH MEASUREMENT - SL = standard; FL = fork.

AGE MEASUREMENT - SC = scale; OT = otolith.

FISHERY - BC = bait catch; TI = trawl incidental; TE = test; CO = commercial; SU = subsistence.

GEAR - Type of gear used to collect samples. Obtain from the code guide (Figure 2A-B).

MESH SIZE - If gill net used to collect samples indicate actual mesh size in inches.
00 = dropout (fish sampled from unknown mesh size).

NET LENGTH - Record length of net in fathoms.

SET/SAMPLE # - Number assigned test fish sets only. Number sequentially starting with 1.
Use a separate numbering sequence for each district and section.

TIME SET - Actual time test net set. Use 24 hour (military) time format. For test fishing only.

HOURS FISHED - Total hours (in tenths) test net fished.

PAGE - Number sequentially starting with 1. Use a separate numbering sequence for each district, section and gear type. Be sure this number corresponds to the correct scale slide number.

LENGTH - Record all lengths in millimeters.

WEIGHT - Record all weights in grams.

OBTAIN THE FOLLOWING INFORMATION FROM THE CODE GUIDE (Figure 2A-B).

DISTRICT	SEX
SECTION	GONAD
LOCATION	AGE

Figure 1C.

AWL CODES

Gear
1 = Variable mesh gillnet - floating
2 = Variable mesh gillnet - sinking
3 = Set gillnet
4 = Drift gillnet
5 = Purse seine
6 = Beach Seine
7 = Otter trawl
8 = Hand picked
9 = Dip net

Sex
1 = Male
2 = Female
3 = Juvenile
4 = Unknown

Speciman Age
1-17 = actual fish age in years
18 = regenerated
19 = illegible
20 = missing

Mesh Size
1-4 7/8 = inches
00 = dropout

GONADS (Relative Maturity)

Maturity	Key Characteristics
1	Virgin herring. Gonads very small, threadlike, 2-3 mm broad. Ovaries wine red. Testes whitish or grey brown.
2	Virgin herring with small sexual organs. The height of ovaries and testes about 3-8 mm. Eggs not visible to naked eye but can be seen with magnifying glass. Ovaries a bright red color; testes a reddish grey color.
3	Gonads occupying about half of the ventral cavity. Breadth of sexual organs between 1 and 2 cm. Eggs small but can be distinguished with the naked eye. Ovaries orange; testes reddish grey or greyish.
4	Gonads almost as long as body cavity. Eggs larger varying in size, opaque. Ovaries orange or pale yellow; testes whitish.
5	Gonads fill body cavity. Eggs, large round; some transparent. Ovaries yellowish, testes milkwhite. Eggs and sperm do not flow, but sperm can be extruded by pressure.
6	Ripe gonads; eggs transparent; testes white; eggs and sperm flow freely.
7	Spent herring. Gonads baggy and bloodshot. Ovaries empty or containing only a few residual eggs. Testes may contain remains of sperm.
8	Recovering spents. Ovaries and testes firm and larger than virgin herring in Stage II. Eggs not visible to naked eye. Walls of gonads striated; blood vessels prominent. Gonads wine red color; (this stage passes into Stage III).

Figure 2 A.

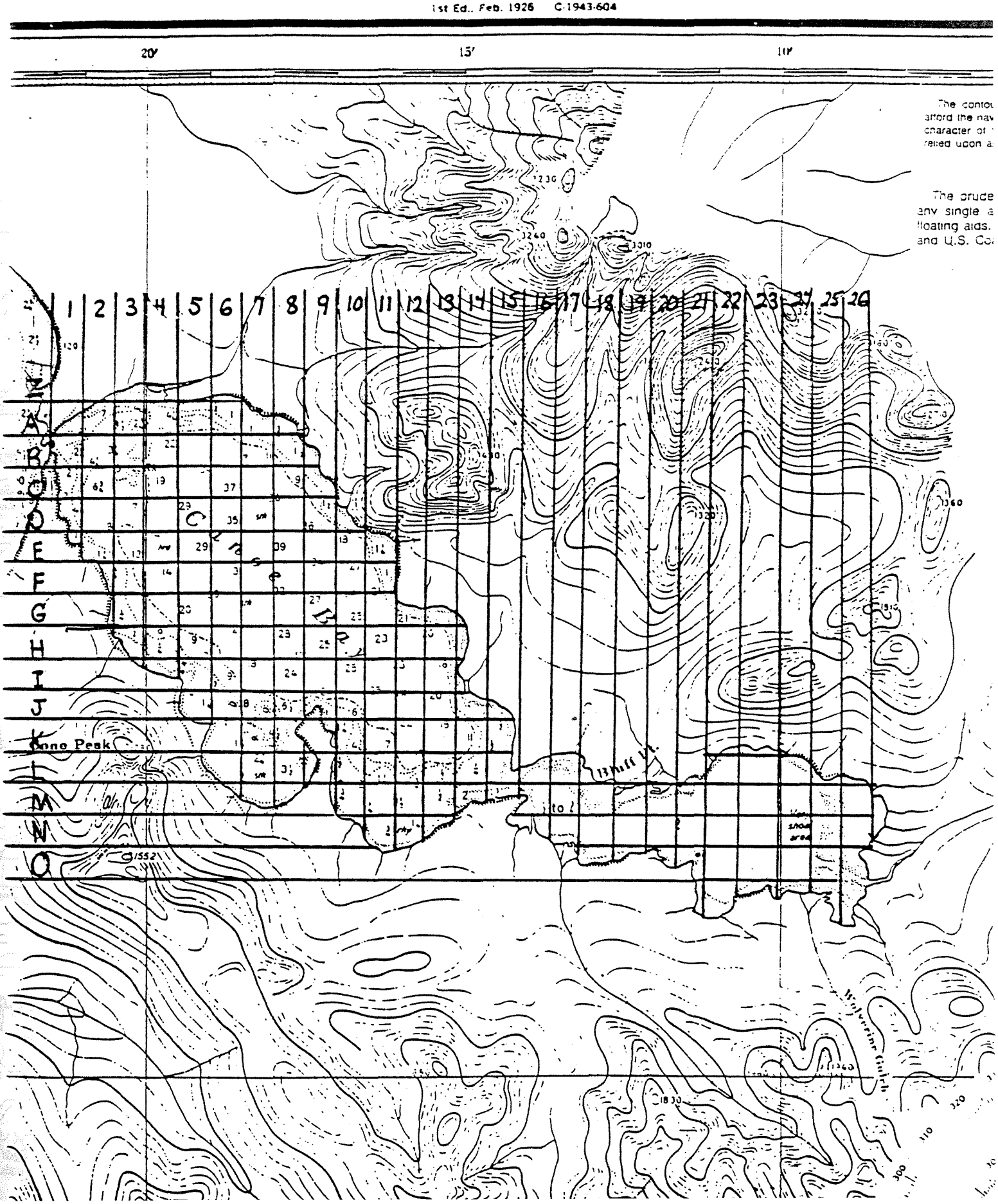


Figure 2 B.

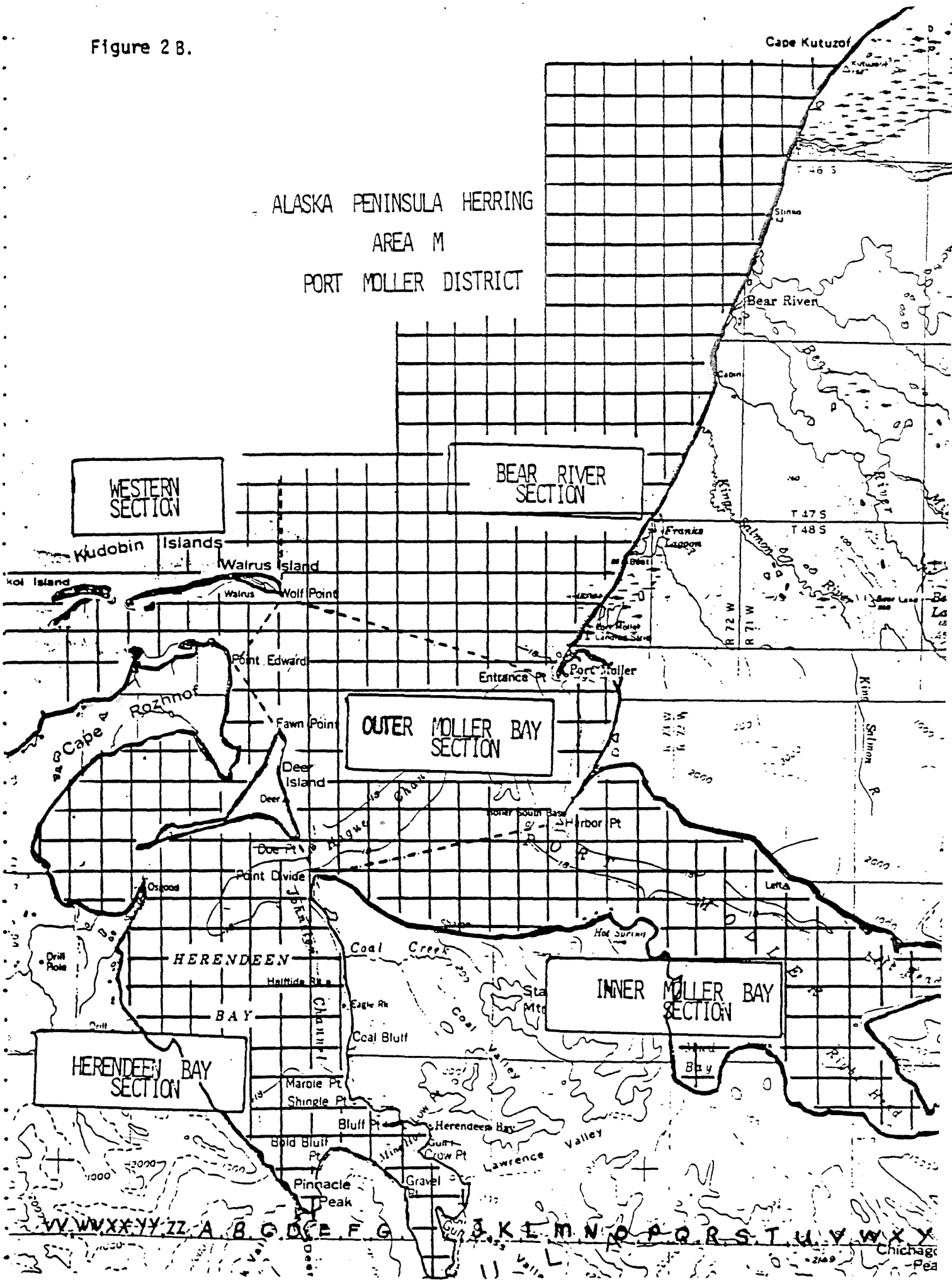
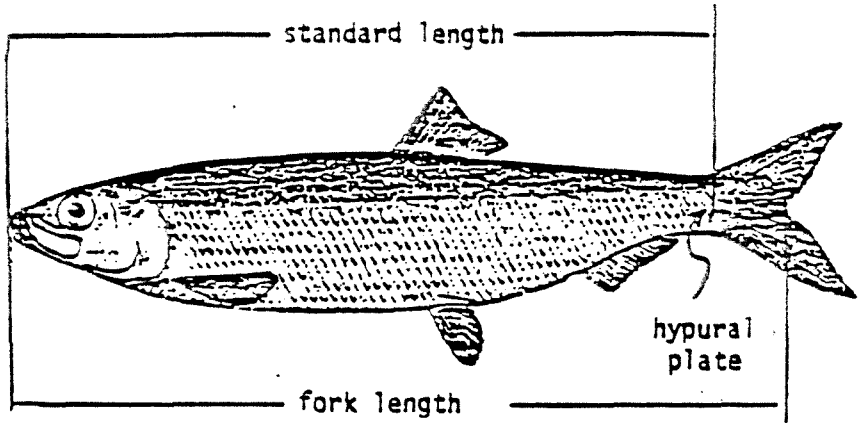
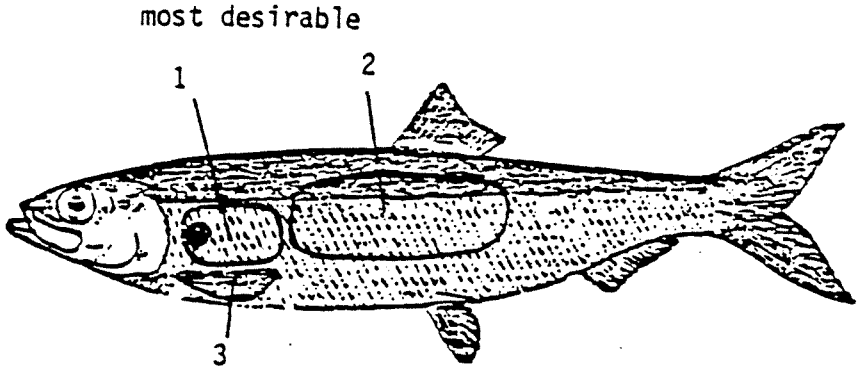

ALASKA PENINSULA HERRING
AREA M
PORT MOLLER DISTRICT
WESTERN SECTION
BEAR RIVER SECTION
OUTER MOLLER BAY SECTION
INNER MOLLER BAY SECTION
HERENDEEN BAY SECTION
Cape Kutuzof
Bear River
Kudobin Islands
Walrus Island
Wolf Point
Point Edward
Entrance Pt
Port Moller
Cape Roznnof
Fawn Point
Deer Island
Harbor Pt
Doe Pt
Point Divide
Osgood
HERENDEEN BAY
Coal Creek
Coal Bluff
Marble Pt
Shingle Pt
Bluff Pt
Herendeen Bay
Bold Bluff Pt
Crow Pt
Pinnacle Peak
Gravel Pt
Lawrence Valley
Franks Lagoon
Kings Salmon River
T 46 S
T 47 S
T 48 S
R 72 W
R 71 W
VV WW XX YY ZZ A B C D E F G J K L M N O P Q R S T U V W X Y

Figure 3.

Standard Length: (Use) the straight line distance from the anterior most part of the fish, including the lower jaw with the mouth closed, to the end of the vertebra (hypural plate). The vertebra end is recognized by the folding of the skin on an unskinned fish when the tail is sharply bent.



Fork Length: (Don't Use) the straight line distance from the anterior most portion of the snout or upper jaw to the extreme end of the center of the caudal fin. It does not include a projectin lower jaw.

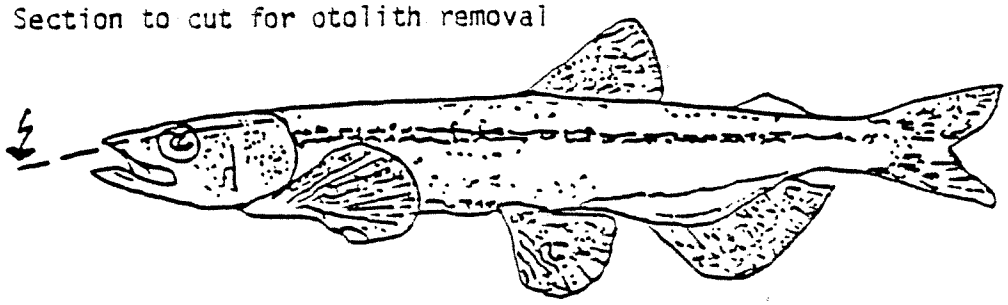


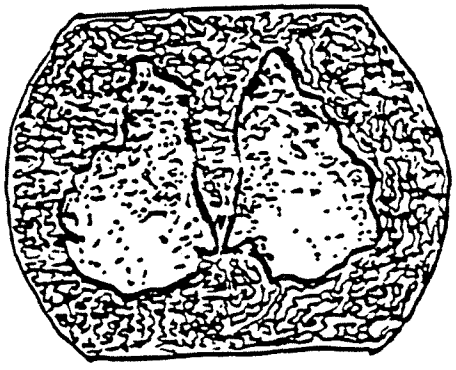
(Under pectoral fin, odd shape - but okay)

Preferred body areas from which to collect scales for aging are shown above. Scales should be collected from the left side of the herring if possible. If no scales are present in any of the above areas on the left side, check the right side (using the same preference sequence). If scales are really scarce, take any good one you can find that is not regenerated.

Figure 4.

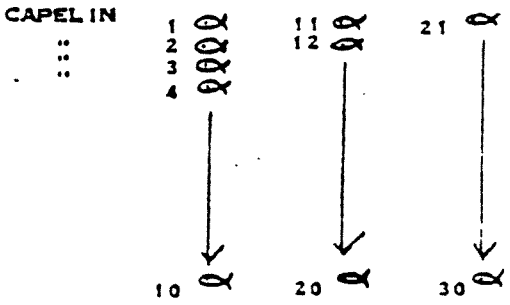
Section to cut for otolith removal





(Actual size)

Capelin Otoliths



CAPELIN SHOULD BE POSITIONED SO AS TO MATCH ENTRIES ON AWL FORMS. THIS IS IMPORTANT FOR LATER SCALE SAMPLING.

CONFIDENTIAL

STATE/FEDERAL NATURAL RESOURCE DAMAGE ASSESSMENT
DETAILED STUDY PLAN

DRAFT

Project Title:	EFFECTS OF HYDROCARBONS ON BIVALVES
Study ID Number:	Fish/Shellfish Study Number 13
Lead Agency:	State of Alaska, ADF&G; Commercial Fish Division
Cooperating Agency(ies):	Federal: USFS State: DNR
Principal Investigator:	Alan S. Davis, Fishery Biologist
Assisting Personnel:	Wayne Donaldson, Fishery Biologist Charlie Trowbridge, Fishery Biologist Peggy Murphy, Biometrician Two Fisheries Technicians
Date Submitted:	October 12, 1989

	Signature	Date
Principal Investigator:	[signature]	[illegible]
Supervisor:	[signature]	[illegible]
OSIAR Senior Biometrician:		
OSIAR Program Manager:	[signature]	10/17/89
OSIAR Director:	[signature]	10/17/89

INTRODUCTION

The goal of this project is to determine whether the Exxon-Valdez oil spill will have a measurable impact on populations of bivalve mollusks in Prince William Sound. Bivalve mollusks are an important component of the food chain, and they support subsistence and sport fisheries in Prince William Sound. Because they are relatively sedentary and occupy nearshore areas, bivalves may be particularly susceptible to contamination by oil. It is hypothesized that increased hydrocarbons in nearshore sediments could affect bivalves by increasing mortality, decreasing growth, or causing sublethal injuries. This study seeks to evaluate these potential effects by comparing data obtained from several beaches representing a range of oil contamination. Data on growth obtained in 1989 will provide a baseline for comparison with growth data to be taken at a future date. Documenting effects on clams, cockles, and other species is required to advise the public of the full scope of impact by the oil spill on current and future employment, recreation, and lifestyles of coastal communities on Prince William Sound.

OBJECTIVES

1. Test the hypothesis that the level of hydrocarbons in bivalves is not related to the level of oil contamination of a beach. The experiment is designed to detect a difference of 1.9 standard deviations in hydrocarbon content with the probability of making a type I and type II error of 0.05 and 0.1, respectively.

2. Test the hypothesis that the level of hydrocarbons in sediments is not related to the level of oil contamination of a beach. The experiment is designed to detect a difference of 1.9 standard deviations in hydrocarbon content with the probability of making a type I and type II error of 0.05 and 0.1, respectively.

3. Document the presence and type of damage to tissues and vital organs of bivalves sampled from beaches with no, intermediate, and high levels of oil contamination such that differences of ±5% can be determined between impact levels 95% of the time.

4. Test the hypothesis that the proportion of dead bivalves sampled on beaches is not related to the level of oil contamination of a beach. The experiment is designed to detect a difference of 5% in the proportion of live to dead bivalves from beaches with no, intermediate, and high levels of oil contamination with the probability of making a type I and type II error of 0.05 and 0.1, respectively.

5. Test the hypothesis that the growth rate of littleneck clams is the same at beaches of no, intermediate, and high levels of oil impact. This experiment was designed to detect a difference in mean shell height equal to the difference between the mean shell height at age i and age i+1 clams with the probability of making a type I error equal to 0.01 and probability of making a type II error equal to 0.05.

6. Identify potential alternative methods and strategies for restoration of lost use, populations, or habitat where injury is identified.

METHODS/DATA ANALYSIS

This project will be conducted in two phases. Phase I is a complete sampling of all the species and beaches listed here for hydrocarbon and necropsy samples. Growth information will be collected only for the most common species, littleneck clams. During Phase II, a heavily oiled beach will be monitored periodically to determine presence of abnormal numbers of dead clams. If abnormal numbers of dead clams are found, the entire circuit of beaches described here will be sampled again. In the second round of sampling only littleneck clams will be collected for necropsy and hydrocarbon analysis.

Study Sites

Beaches known to contain clams were surveyed to determine the level of oil contamination. Nine study sites in Prince William Sound representing three levels of oil contamination (subjectively rated as no, intermediate, or high contamination) were chosen to be sampled (Table 1, Figure 1). Beaches with no oil contamination are Hell's Hole, Pellew Cove, and Simpson Bay. Beaches with light or intermediate oil contamination are Ellamar, Outside Bay, Gibbon Anchorage. Beaches with heavy oil contamination are Snug Harbor, Wilson Bay, and Fox Farm (Elrington Is.).

Table 1. Study beaches representing three levels of oil impact.

IMPACT LEVEL		
NONE	INTERMEDIATE	HIGH
Hell's Hole	Ellamar	Snug Harbor
Pellew Cove	Outside Bay*	Wilson Bay
Simpson Bay	Gibbon Anchorage	Fox Farm

* Sites sampled by NMFS mussel studies during 1977-1980 and 1989-1990.

For each sample site the following site description information will be recorded (Appendix A, Form 1 and Form 2): site orientation (N-NW etc.), latitude, longitude, beach slope, low tide height, percent dominant substrate composition, temperature and salinity of the water, weather and wave action. Temperature and salinity of the water will be measured at a distance of approximately 5 meters offshore from the sampled beach at the daily low slack tide.

Sample Design

Beaches will be sampled at maximum low tides for a monthly tidal cycle. At each beach, three sampling transects will be run to insure complete coverage of the beaches as distribution of oil on the beaches is unknown. Transects will be perpendicular to the water's edge and parallel to each other with a total distance between the each transect of 15 meters (Appendix B). Transects were perpendicular to the water to insure complete sampling of clam habitat. The top of each transect is placed at the +1.6 meter tide level and the bottom of the transect at the lowest tide level. Tidal height will be determined using reference points from a standard tide table, a hand level, and a stadia rod. Transect number, bottom of the transect tidal height, top of the transect tidal height, middle of the transect tidal height, and distance from the top of the transect to each remaining sampling quadrate will be recorded.

Prior to sampling, the upper distribution of clams and cockles will be determined by removing sediment to a depth of 30 cm (12 in) along a trench adjacent to the proposed transect. The trench is dug starting from the top of the transect and continuing until clams and cockles are encountered.

A total of seven quadrates will be sampled from each transect to obtain hydrocarbon and necropsy specimens. Sample quadrates are each 0.5 m^2. Additional sampling or complete sampling of each transect (all possible sampling quadrates) may be necessary if insufficient numbers of clams or cockles are recovered within the seven sampling quadrates to meet project objectives. Quadrates will be sampled from the top to the bottom of each transect as the tide recedes. The distribution of clams will extend below the low tide levels encountered during each sampling event. However, the bottom of each transect and the bottom sampling quadrate will occur at the daily low tide level.

The first sample quadrate will be located where the first clam or cockle was encountered in the preliminary trench. The second quadrate will be located equal distance from the top and bottom sampling quadrates. The tidal height of the second quadrate is calculated as half the difference between the tidal height of the first quadrate and the low tide height subtracted from the tidal height of the first quadrate. The third quadrate is taken at an equal distance from the first and second quadrates (a tidal height

approximately one quarter the difference between the tidal height of the first quadrate and the low tide height subtracted from the tidal height of the first quadrate). The fourth sample is taken at a point equal distance between the second and third sample quadrates. The fifth sample is taken at a point equal distance between the low tide level and the second quadrate, a tidal height equal to three quarters the difference between the tidal height of the first quadrate and the low tide height subtracted from the tidal height of the first quadrate. The sixth sample is taken in the area equal distance between the fifth and second sample quadrates. The last sampling quadrate is placed at the low tide level (Appendix B).

Sediment Sample Collection for Hydrocarbon Analysis

A total of three sediment samples will be collected from each beach site (one from each transect). All sediment samples will be collected before bivalve sampling is performed. The hydrocarbon sample from each transect will be a composite sediment sample which will be collected by scooping one tablespoon of sediment to a depth of 2 to 3 cm from each of the seven sample quadrates on a transect.

All samples from each transect will be placed in an 8 oz glass jar rinsed with methylene chloride. Each jar will be labelled with the site name, latitude, longitude, date, "SEDIMENT", transect number, sample number, names of the sampling team members, "BIVALVE", and "ADF&G".

Three composite sediment samples (one per transect) will be obtained from each beach sampled. This will provide a total of 9 samples for each impact level (1 hydrocarbon sample/transect * 3 transects/site = 3 hydrocarbon samples/site; 3 hydrocarbon samples/site * 3 sites/impact level = 9 hydrocarbon samples/impact level).

The small sub-samples of sediment taken from each sampling quadrate will provide a representative mixture of sediment composition and contamination throughout the transect. One composite sediment sample for each transect at each site provides 9 composite samples for each impact level (no, intermediate, and high). The industry standard is 8 samples for each treatment level. A sample size of 9 composite samples is considered an adequate number of samples to detect a difference in sediment contamination between impact levels at the desired α and β levels.

Bivalve Sample Collection

During Phase I, one species of cockle Clinocardium nutalli and two common species of clam, littleneck clam Protothaca staminea and butter clam Saxidomus giganteus, will be sampled for hydrocarbon analysis, necropsy, and to estimate percent of live and dead clams. Littleneck clams will also be collected to estimate age and growth statistics. During Phase II, only the most common species,

littleneck clam, will be sampled for comparison with Phase I results.

Hydrocarbon Samples:

Specimens for hydrocarbon analysis will be taken from each sampling quadrates before any other specimen sampling is conducted. Bivalves of each species will be randomly selected for hydrocarbon analysis from sampling quadrates at each site. Care will be taken to avoid contamination of a specimen by sediments not immediately surrounding the specimen. Each sample will be placed directly in a sample container before another bivalve is obtained.

One hydrocarbon sample for each species will be obtained from each transect. For littleneck clams and butter clams, each hydrocarbon sample will be composed of 14 specimens. The 14 samples from each transect (1 hydrocarbon sample) will be selected by randomly picking two clams from each of the seven sampling quadrates. Each clam must have a shell length of 2-5 cm.

Each hydrocarbon sample for cockles will be composed of six individuals. The six cockles from each transect will be selected by randomly picking six of the seven sampling quadrates then randomly selecting one cockle (size 5-7 cm) from each of the sampling quadrates.

Bivalve samples are being limited to a particular size range because rates of uptake, metabolism, and depuration by clams and cockles probably change with size. If specimens of the desired size are not found in each of the sampling quadrates, then the desired number of additional specimens will be collected from the other sample quadrates.

Specimens of each species from each transect will be placed together in a non-plastic container and the air will be evacuated from the container. A 16 oz squat jar should be large enough for 14 littleneck clams and 14 butter clams. A 32 oz jar may be large enough to hold 6 cockles. Alternatively, clams and cockles can be wrapped in paper towels then wrapped in aluminum foil. The foil packages can be stored in metal coffee cans covered with aluminum foil prior to sealing with the plastic lid. All containers and pieces of aluminum foil must be rinsed with methylene chloride before they are used. Each container will be labelled with the site name, latitude, longitude, date, species, transect number, names of the sampling team members, "BIVALVE" and "ADF&G". All hydrocarbon samples will be stored at
-40° F until hydrocarbon analysis is initiated.

Combined tissue samples from each sampling quadrate will provide a representative mixture of bivalve tissue composition and contamination throughout the transect. The desired size of each composite tissue sample is 15 gm. The number of bivalves to provide this sample from each transect was estimated based on the average

size of individuals of each species. An estimate of 3 hydrocarbon samples from each site is needed for detecting contamination between impact levels (Dr. B. Clark, personal communication). A sample size of 9 composite samples per impact level will allow the detection of differences in hydrocarbon content of 1.9 standard deviations with α and β levels of 0.05 and 0.1, respectively.

Necropsy Samples:

Collection of specimens for necropsy will begin only after all hydrocarbon samples have been taken. Total sample size is 20 live or moribund specimens of each species from each beach site. With 20 bivalves sampled from each beach, the total sample for each treatment (no, intermediate, and high oil contamination) will be 60. This sample size will allow detection of differences in presence of tissue damage of ±5% with 95% confidence between samples obtained from beaches with different levels of oil impact (Dr. Ted Meyers, Alaska Department of fish and game, personal communication). This sample size will allow detection of gross differences between beaches with no, medium, and high oil impact.

One specimen of each species will be randomly selected from each sampling quadrate. This will yield a total of 21 specimens. One specimen from the 21 collected will be randomly selected and discarded from the sample to achieve a sample size of 20 specimens. Sampling procedures and quality assurance will be conducted as outlined in the histopathology guidelines, and sample preparation will be followed as outlined in Appendix C. Histopathological analysis of bivalve tissues will include all criteria listed in the histopathology guidelines. Necropsies will be performed by a qualified contractor approved by the Histology Technical Group.

Bivalve Mortality Samples:

All live and recently dead specimens of each species from each sampling quadrate will be separated from the sediment and placed in containers. Recently dead bivalves are defined as those bivalves with tissue remaining attached to the shell. Each container will be identified with the species name, transect number, and sampling quadrate number. Each container will be set aside until quadrate sampling is completed. After all quadrates in each transect have been sampled, live specimens and recently dead specimens will be counted and placed in separate containers. Numbers of live and dead bivalves will be recorded by sampling quadrate and transect on Form 4 (Appendix A).

An additional 30 meter transect located along the high tide line above the sampling stations will be sampled. All recently dead bivalves along this transect will be counted.

Growth and Age Samples:

Only littleneck clams will be collected for growth and age estimation. A total of 100 specimens will be collected from each transect at each site. From each transect five sampling quadrates will be selected at random. From each of these, 14 specimen will be randomly sampled from the quadrate containers. Fifteen specimens will be randomly sampled from the remaining two quadrate containers. The specimens from each sampling quadrate container will be placed in separate bags. Each bag will be labelled with the site name, latitude, longitude, date, transect number, sampling quadrate, names of the sampling team, "BIVALVE" and "ADF&G". Specimens from each sampling quadrate will be cooked and shucked at a later time. Once specimens have been cooked and shucked, each valve for each specimen is labeled with a specimen number unique to that specimen. Care will be taken to keep specimens from different sampling quadrates separate until specimen numbers have been assigned and recorded for each sampling quadrate.

The sample of 100 specimens from each transect will provide 300 samples from each beach and 900 clams for each level of beach impact. Sample size for growth is based on the difference between mean shell height for age i and age i+1 clams, variance in shell height for age i+1 clams, probability of making a type I error equal to .01 and probability of making a type II error equal to .05 (Netter and Wasserman 1985). Data for mean shell height and variance in shell height was taken from Paul and Feder (1973). Sample size for detecting difference in growth at age of clams between impact levels was estimated at 261-275 for each impact level (Appendix D). This sample size was rounded up to 300 clams. The purpose of 3 sites for each impact level is to provide replicates at each impact level.

Data Analysis

To address objective 2 (hydrocarbons in sediments), an analysis of variance will be used to test for differences in hydrocarbon content in sediment between sites. Differences in sediment hydrocarbon content will verify that control sites (areas of no oil impact) are in fact "controls". These differences will also permit post-stratification of sample sites according to level of impact.

To address objective 1 (hydrocarbons in bivalve tissues), an analysis of variance will be performed on the hydrocarbon content of clam and cockle samples among sites. The results of this test will be related to the level of sediment impact.

To address objective 4, the proportion of dead clams among sites will be subjected to analysis of variance, and the results related to level of oil contamination of sediments. Statistical differences in injury rates (within specific categories of injury as outlined in the histology guidelines) between impact areas will be evaluated using chi-square analysis (Objective 3).

To provide baseline (pre-impact) information on variance in growth at age among sites, an analysis of variance on growth parameters from clams between areas will be conducted. Growth parameters will be determined for various growth curves, such as Gompertz, von Bertalanffy, or polynomial equations. Growth parameters will be presented for the most appropriate growth models only. These beach sites will be resampled after 4 years. An analysis of variance on growth parameters obtained from fitting algorithms for clam growth after impact (1990-1994) will be compared to growth parameters for clam growth prior to impact (approximately 1979-1989) to resolve impact of oil contamination on growth (Objective 5). Graphics will be used to display differences in growth among areas over time, including growth curves (size at age) and growth increment at age by year for each beach.

Statistics for analysis of variance will also be computed for comparisons of hydrocarbon content and proportions of dead clams and cockles. Appropriate statistics for non-linear and polynomial curve fitting will be computed to evaluate the strength of the fit, including mean square, sum of squares, degrees of freedom, etc. Variances will be estimated for all means computed, such as mean growth at age, mean proportion dead, etc.

To address objective 6, all data will be analyzed to determine degree of damage to stocks. Appropriate suggestions will be made for restoration or mitigation measures. This may include restrictions on human usage to reduce mortality rates or may include the need for continued monitoring of stocks.

SCHEDULES AND REPORTS

Date(s)	Activity
April-May 1989	Phase I field sampling. A different beach will be sampled each day.
May-August 1989	Phase II field sampling. During the monitoring phase a single beach will be sampled bi-weekly. If high rates of mortality are determined, all study sites will be resampled.
August-September 1989	Laboratory work, hydrocarbon analysis, etc.
September-December 1989	Data entry and analysis
December 1989	Preliminary report on impacts of oil on clams (will not include pre- and post-impact comparisons for growth.

PROJECT BUDGET

Line Item	Category	Budget
100	Personnel Services	$ 20.5
200	Travel	$ 3.5
300	Contractual	$ 50.5
400	Commodities	$ 8.7
500	Equipment	$ 3.0
700	Grants	$ 0
Total		$ 86.2

FUNDED PERSONNEL

Class	PCN	PFT_mm	SFT_mm
FB III	11-1084	2	
FB I	11-1641	2	
FT II	11-n-		2

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Clark, B. Personal communication. National Marine Fisheries Service, Environmental Conservation Division, 2725 Montlake Boulevard East, Seattle, WA.

Meyers, T. Personal communication. Alaska Department of Fish and Game, Fisheries Rehabilitation and Economic Development Division, P.O. Box 3-2000, Juneau AK.

Neter, J., W. Wasserman and M. Kutner. 1985. Applied Linear Statistical Models. Richard D. Irwin, Homewood Illinois.

Paul, A.J. and H.M. Feder. 1973. Growth, recruitment, and distribution of the littleneck clam, Protothaca staminea in Galena Bay, Prince William Sound, Alaska. Fishery Bulletin 71(3):665-677.

Shirley, T. Personal communication. University of Alaska, Southeast. School of Fisheries and Ocean Sciences, 11120 Glacier Highway, Juneau, AK.

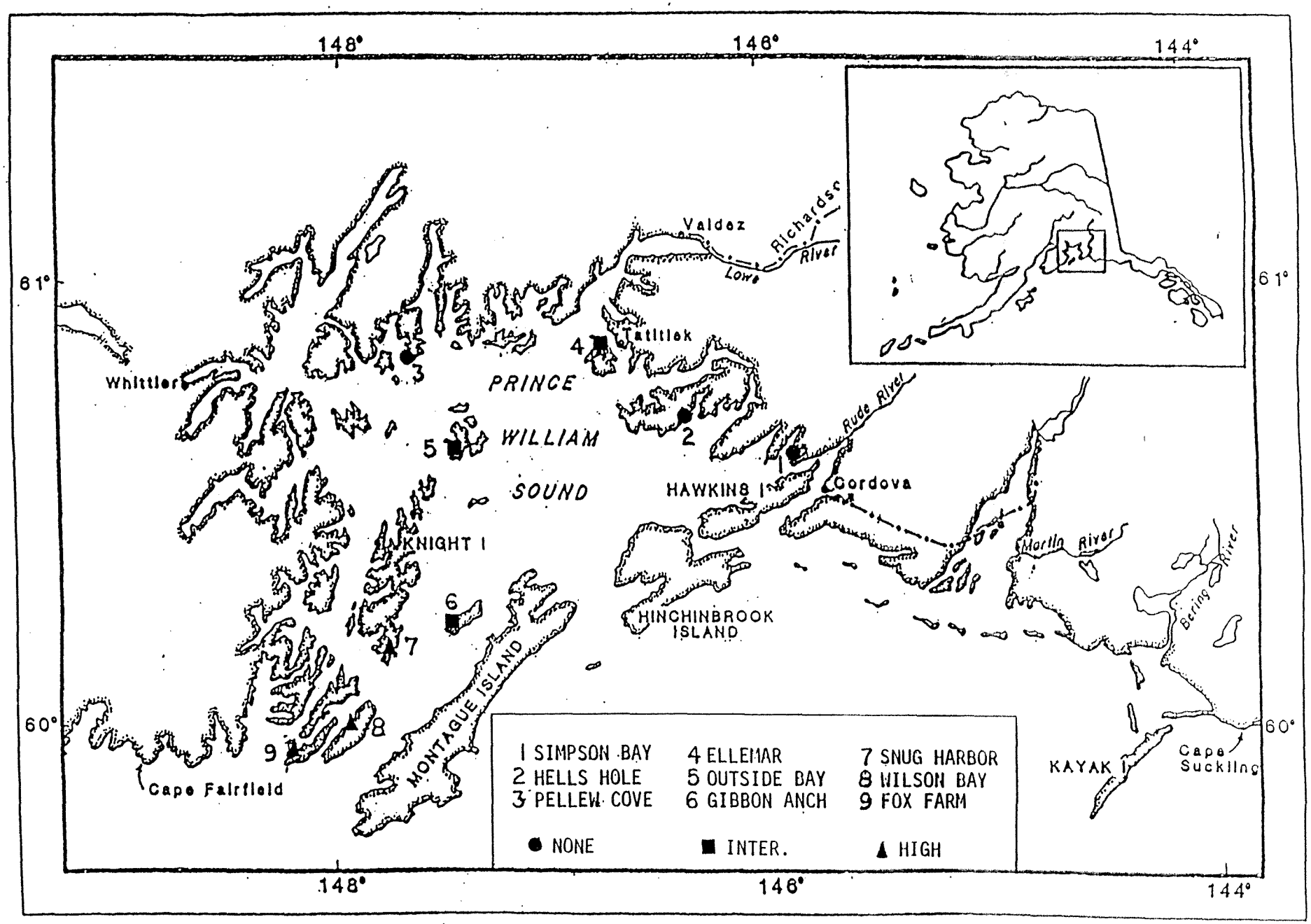

148°
146°
144°
61°
60°
Valdez
Richardson
River
Lowe
Whittier
Latitlek
PRINCE
WILLIAM
SOUND
KNIGHT I
HAWKINS I
Cordova
Rude River
Martin River
Bering River
HINCHINBROOK
ISLAND
MONTAGUE ISLAND
Cape Fairfield
KAYAK I
Cape Suckling
1 SIMPSON BAY
2 HELLS HOLE
3 PELLEW COVE
4 ELLEMAR
5 OUTSIDE BAY
6 GIBBON ANCH
7 SNUG HARBOR
8 WILSON BAY
9 FOX FARM
NONE
INTER.
HIGH

Figure 1

ADF&G BIVALVE OIL PROJECT SITE DESCRIPTION FIELD FORM

Site Name _____ Date _____

Latitude _____ Longitude _____

Skipper Name _____

Vessel Name _____ Vessel Ak Number _____

Vessel Description _____

Sampling Team	Agency

Waves Code _____ Description _____

Weather Code _____ Description _____

Air Temp _____ Sea Temp _____ Salinity _____

Beach Substrate % _____ Code _____ Description _____

Beach Slope _____ Beach Orientation (eg N-NW) _____

NOAA Reference Point _____ NOAA Low Tide Height _____

Film Role Number _____ Photograph numbers _____ _____

Waves

Code	Description
1	Glassy
2	Rippled
3	Wavelets
4	Slight 2-4'
5	Moderate 4-8'
6	Rough 8-13'
7	Very Rough 13-20'

Weather

Code	Description
1	Clear
2	Partly Cloudy
3	Overcast
4	Fog or Thick Haze
5	Showers
6	Squalls
7	Drizzle
8	Rain
9	Rain and Snow
10	Snow
11	Blizzard

Substrate

Code	Description
1	Mud-Silt
2	Clay
3	Sand
4	Granule (2-4mm
5	Pebble(4mm-3cm
6	Rock Fragments (3-6cm)
7	Cobble Shingle (6-15 cm)
8	Rock (15-25 cm
9	Boulder(>25 cm

ADF&G BIVALVE OIL PROJECT
TRANSECT DESCRIPTION FORM 2

Site Name _____ Date _____

Latitude _____ Longitude _____

	Transect 1	Transect 2	Transect 3
Bottom of Transect Tidal Height			
Top of Transact Tidal Height			
Middle of Transect Tidal Height			
Length of Transect to Quadrant 3			
Length of Transect to Quadrant 4			
Length of Transect to Quadrant 5			
Length of Transect to Quadrant 6			
Total Length of Transect			

Distance Between Top of Transect 1 and 2: _____ 2 and 3: _____

Total Width of Sampling Site _____

	Total Number of Clam Shells	Number of Recently Dead Clam Shells
High Tide Line 30 Meter Transect		

SPEDIMENT AND SPECIMEN FORM 3

Site Name _____ Date _____

Latitude _____ Longitude _____

SAMPLE COLLECTION - CHECK THE BOX IF THE SAMPLE WAS COLLECTED

	Transect 1		Transect 2		Transect 3	
SEDIMENT HYDROCARBON	Yes	#Q	Yes	#Q	Yes	#Q
composite sediment sample 1 & number of quadrants sampled	☐	_	☐	_	☐	_
composite sediment sample 2 & number of quadrants sampled	☐	_	☐	_	☐	_
composite sediment sample 3 & number of quadrants sampled	☐	_	☐	_	☐	_

	Transect 1			Transect 2			Transect 3		
LITTLENECK CLAM HYDROCARBON	Yes	#S	#Q	Yes	#S	#Q	Yes	#S	#Q
composite hydrocarbon sample, number of specimens sampled, number of quadrants sampled & size range	☐	_	_	☐	_	_	☐	_	_
	_____			_____			_____		
BUTTER CLAM HYDROCARBON composite hydrocarbon sample, number of specimens sampled, number of quadrants sampled & size range	☐	_	_	☐	_	_	☐	_	_
	_____			_____			_____		
COCKLE HYDROCARBON composite hydrocarbon sample, number of specimens sampled, number of quadrants sampled & size range	☐	_	_	☐	_	_	☐	_	_
	_____			_____			_____		

	Transect 1			Transect 2			Transect 3		
LITTLENECK CLAM NECROPSY	Yes	#S	#Q	Yes	#S	#Q	Yes	#S	#Q
composite necropsy sample, number of specimens sampled, number of quadrants sampled & size range	☐	_	_	☐	_	_	☐	_	_
	_____			_____			_____		
BUTTER CLAM NECROPSY composite necropsy sample, number of specimens sampled, number of quadrants sampled & size range	☐	_	_	☐	_	_	☐	_	_
	_____			_____			_____		
COCKLE NECROPSY composite necropsy sample, number of specimens sampled, number of quadrants sampled & size range	☐	_	_	☐	_	_	☐	_	_
	_____			_____			_____		

ADF&G BIVALVE OIL PROJECT
CLAM MORTALITY FORM 4

Site Name _____ Date _____

Latitude _____ Longitude _____

	Transect 1		Transect 2		Transect 3	
	# Alive	# Dead	# Alive	# Dead	# Alive	# Dead
SAMPLING QUADRANT 1						
Littleneck clam	___	___	___	___	___	___
Butter clam	___	___	___	___	___	___
Cockle	___	___	___	___	___	___
SAMPLING QUADRANT 2						
Littleneck clam	___	___	___	___	___	___
Butter clam	___	___	___	___	___	___
Cockle	___	___	___	___	___	___
SAMPLING QUADRANT 3						
Littleneck clam	___	___	___	___	___	___
Butter clam	___	___	___	___	___	___
Cockle	___	___	___	___	___	___
SAMPLING QUADRANT 4						
Littleneck clam	___	___	___	___	___	___
Butter clam	___	___	___	___	___	___
Cockle	___	___	___	___	___	___
SAMPLING QUADRANT 5						
Littleneck clam	___	___	___	___	___	___
Butter clam	___	___	___	___	___	___
Cockle	___	___	___	___	___	___
SAMPLING QUADRANT 6						
Littleneck clam	___	___	___	___	___	___
Butter clam	___	___	___	___	___	___
Cockle	___	___	___	___	___	___
SAMPLING QUADRANT 7						
Littleneck clam	___	___	___	___	___	___
Butter clam	___	___	___	___	___	___
Cockle	___	___	___	___	___	___

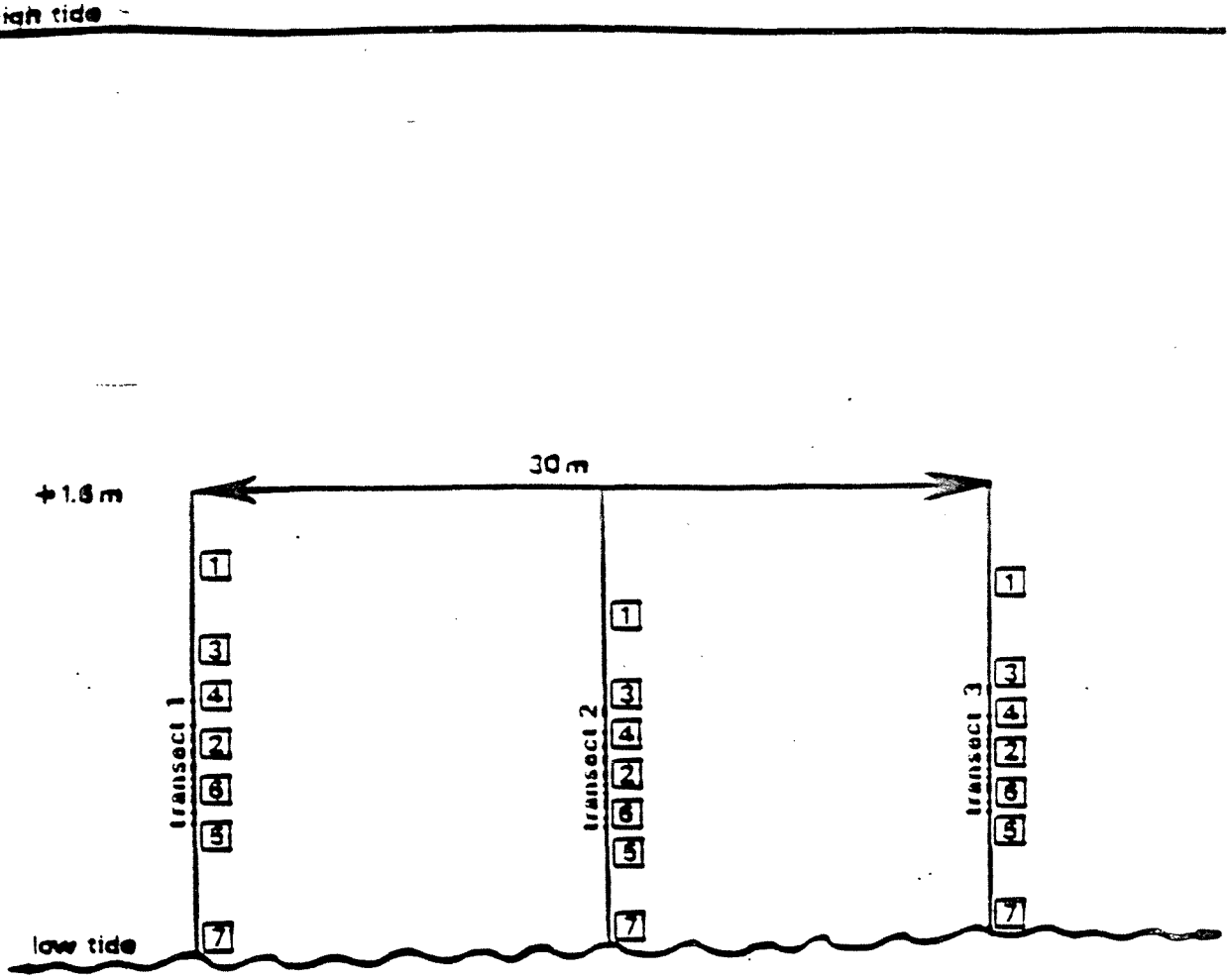
ADF&G BIVALVE OIL PROJECT
LITTLENECK CLAM GROWTH AND AGE FORM 5

Site Name _____ Data _____

Latitude _____ Longitude _____

	Transect 1	Transect 2	Transect 3
QUADRANT 1, number of specimens	_____	_____	_____
& specimen numbers	_____ _____	_____ _____	_____ _____
QUADRANT 2, number of specimens	_____	_____	_____
& specimen numbers	_____ _____	_____ _____	_____ _____
QUADRANT 3, number of specimens	_____	_____	_____
& specimen numbers	_____ _____	_____ _____	_____ _____
QUADRANT 4, number of specimens	_____	_____	_____
& specimen numbers	_____ _____	_____ _____	_____ _____
QUADRANT 5, number of specimens	_____	_____	_____
& specimen numbers	_____ _____	_____ _____	_____ _____
QUADRANT 6, number of specimens	_____	_____	_____
& specimen numbers	_____ _____	_____ _____	_____ _____
QUADRANT 7, number of specimens	_____	_____	_____
& specimen numbers	_____ _____	_____ _____	_____ _____

Appendix 3.



Note - the tidal height of sampling quadrant 1 may vary at each transect.

HISTOLOGICAL SAMPLE PREPARATION FOR BIVALVE MOLLUSCS - ADFG FRED PATHOLOGY

NOTE: Only live or moribund bivalves will be suitable for processing. Histopathological changes caused by toxic chemicals are often very subtle at best. Tissues in dead bivalves autolyze very quickly and will mask these changes. Do not collect and process dead bivalve molluscs.

1. The fixative to be used is Bouin's solution (formula attached).

2. The volume of fixative should be 10 times the volume of the tissue. This is important since any less fixative may result in tissue autolysis and worthless samples.

3. The sample size per site and species will be 30 bivalves, live or moribund.

4. Bivalves less than 6 cm in length (shucked) can be fixed whole by dropping into preservative. Animals must be shucked cleanly from the shell by severing adductor muscles (diagram) prior to fixation. Discard the shell unless there is some type of shell deformity or otherwise abnormal valve. In such a case the shell should be included and attached to the donor animal by wrapping both in gauze.

5. Larger bivalves will need about 3 incisions (anterior, mid, posterior) made across the surface of the animal about midway through the tissues. Do not cut completely through the animal so that individual specimens remain intact and tissues do not become mixed.

6. Tissue and shell abnormalities must be noted on a necropsy field sheet (attached) respectively numbered for a particular animal (bag in gauze and label if necessary). If no abnormalities within the 30 specimens are observed then a single field sheet will suffice for that sample series. The field sheet(s) will also contain the label information below and must accompany the samples in a zip loc bag.

7. A label with bivalve species, size range and life stage, date of sample, location of sample and contact person's name, address and telephone number must be placed within each of the sample jars.

8. Do not mix samples of different species within the same jar of fixative. Each species requires a separate sample jar(s).

9. Place sample jars and zip loc bag containing sample data into a suitable shipping package with adequate packing material to prevent breakage. Plastic jars or containers for fixative and samples work best. Be sure lids are on tight and do not leak.

10. Mail to FRED Fish Pathology Lab: 333 Raspberry Rd., Anchorage 99502 (907-267-2244) or P.O. Box 3-3000, Juneau 99802 (907-465-3577).

11. Notify the fish pathology lab prior to sample shipment so that samples may be expected and tracked en route.

1. Any questions regarding sample preparation should be directed to:

Dr. Ted Meyers
Principal Fish Pathologist III
ADF&G, FRED Division
Juneau Pathology Lab
P.O. Box 3-2000
Juneau, AK 99802 (907) 465-3377

BOUIN'S SOLUTION - FIXATIVE FOR HISTOLOGICAL SAMPLES OF FISH, BIVALVES, AND CRABS- FRED PATHOLOGY, ADFG

1.	Picric Acid, saturated aqueous solution*	750.0 ml
2.	37 - 40 % Formalin	250.0 ml
3.	Glacial acetic acid	50.0 ml

* Dissolve 20 g picric acid into 1000 ml distilled water with the aid of heat. Allow to cool, decant and use the supernatant fluid.

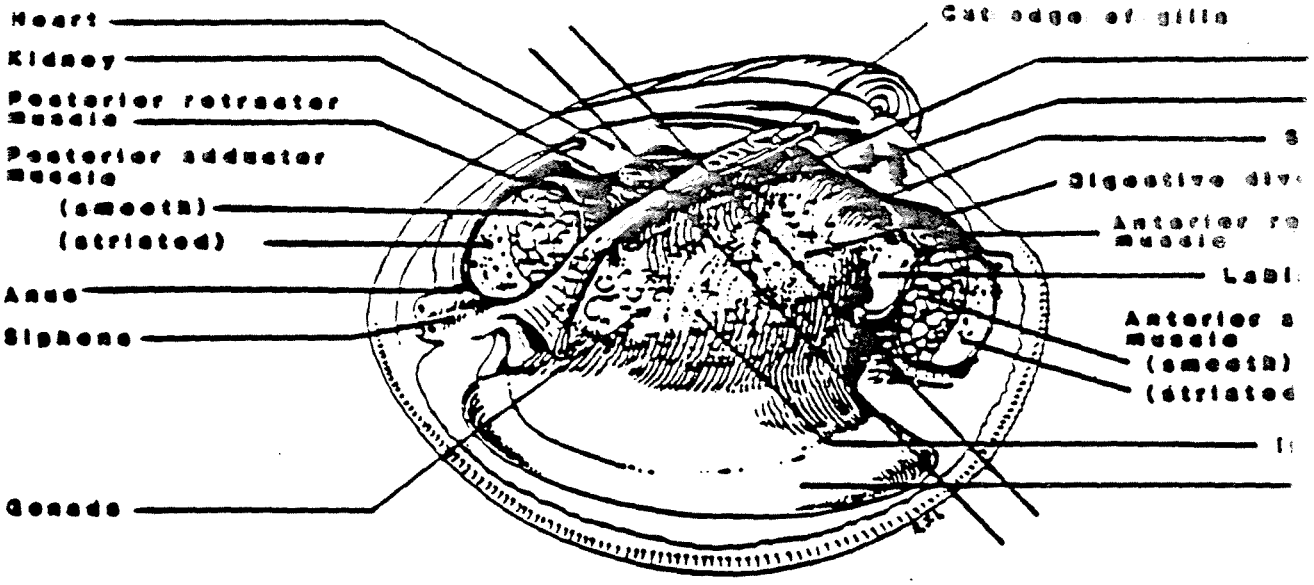


Fig. 44. Gross anatomy of hard clam. The bold parallel lines show the locations where the clams for cross-sections should be taken (Illustration by A.L. Lippson).

NECROPSY FIELD DATA SHEET FOR HISTOLOGICAL SAMPLES, FRED PATHOLOGY, ADF&G

COLLECTOR/ADDRESS/TELEPHONE #

SPECIES

NUMBER SPECIMENS IN SAMPLE

SIZE RANGE

LIFE STAGE

DATE OF COLLECTION

LOCATION OF COLLECTION (SITE NAME OR NUMBER)

ABNORMALITIES OBSERVED PER SPECIMEN NUMBER

Appendix D

Sample size estimation for detecting differences in growth (height) of Protothaca staminea between impacted and non-impacted sites in Prince William Sound

Age	Δ $m_{i+1}-m_i$	σ_{i+1}	Δ/σ	α =.10 $1-\beta$=.90 n	α =.05 $1-\beta$=.95 n	α =.01 $1-\beta$=.95 n
0-1	.84	.33	2.55	5	7	9
1-2	.67	.70	.96	22	32	43
2-3	2.28	.73	3.12	4	5	7
3-4	1.91	.63	3.03	4	5	7
4-5	2.17	1.87	1.16	15-22	21-32	29-43
5-6	3.15	3.26	.97	22	32	43
6-7	5.77	3.99	1.45	11	15	20
7-8	7.07	4.07	1.74	8	12	16
8-9	3.36	1.96	1.71	8	12	16
9-10	3.53	1.83	1.93	7	9	12
10-11	2.15	2.25	.96	22	32	43
11-12	2.94	1.69	1.74	8	12	16
				N=136-143 site	N=194-205 site	N=261-275 site
				N= 45-48 transect	N= 65-68 transect	N= 87-92 transect

Recommended sample size/transect is 100 specimens.

CONFIDENTIAL

STATE/FEDERAL NATURAL RESOURCE DAMAGE ASSESSMENT
DETAILED STUDY PLAN

DRAFT

Project Title:	INJURY TO PRINCE WILLIAM SOUND CRABS
Study ID Number:	Fish/Shellfish Study Number 14
Lead Agency:	ADF&G; Commercial Fish Division (King Crabs) NOAA; (Dungeness Crabs)
Cooperating Agency(ies):	Federal: USFS State: DNR
Principal Investigator:	Charles Trowbridge, Fishery Biologist Charles O'Clair, Fishery Biologist
Assisting Personnel:	John Hilsinger, Fishery Biologist Wayne Donaldson, Fishery Biologist Lincoln Freese, Fishery Biologist Brad Smith, Fishery Biologist One Fishery Technician
Date Submitted:	October 12, 1989

	Signature	Date
Principal Investigator (ADF&G):	Charles E. Trowbridge	10-10-89
Principal Investigator (NMFS):	[signature]	10/13/89
Supervisor: (ADF&G)	Wayne Donaldson	10-10-89
Supervisor: (NMFS)	[signature]	10/12/89
OSIAR Senior Biometrician:		
OSIAR Program Manager:	[signature]	10/17/89
OSIAR Director:	[signature]	10/12/89

INTRODUCTION

Sensitivity to sublethal environmental concentrations of petroleum hydrocarbons has been documented for several species of crabs. Responses of crabs to these concentrations include reduced molting success, early-postmolt autotomy of limbs and behavioral disorders (Karinen and Rice 1974; Krebs and Burns 1977; Malan 1988). These responses may depend on sex or reproductive state (Krebs and Burns 1977; Jackson et al 1981). Dungeness crab have shown impaired chemosensory antennular flicking response when exposed to water contaminated with Prudhoe Bay crude oil (Pearson et al. 1981). Dungeness crabs exposed to oiled sediments have produced fewer larvae with reduced survival (Karinen et al. 1985). Dungeness crabs may be particularly susceptible to oil contamination because they occupy nearshore habitats in protected bays where they frequently burrow into benthic sediments. If oil becomes incorporated in the fine sediments that occur in these bays it persists and can affect crab populations for several years after an oil spill (Krebs and Burns 1977, Boehm et al. 1987).

This project is designed to assess the long and short term damage to the Dungeness and brown king crab resources of Prince William Sound due to the spillage of Prudhoe Bay crude oil by the Exxon Valdez. The Knight Island Passage area, which was severely impacted by the oil, is the primary habitat of brown king crab in Prince William Sound. Small numbers of Dungeness crab are known to exist in the western Sound. Establishing the degree of impact on these two crab resources is required in order to advise the public of the full scope of damage caused by the oil in Prince William Sound. This information is also critical to future public policy decisions on resource development in Alaska.

The benefits of this project are to: (1) provide data on damage to crabs due to the Exxon Valdez oil spill, (2) help determine compensation required for lost crab resources, and (3) Identify potential alternative methods and strategies for restoration of lost use, populations, or habitat where injury is identified.

OBJECTIVES

1. Determine the levels of hydrocarbons in tissues and eggs of Dungeness crabs and relate those levels to concentrations of hydrocarbons in sediments at four oiled and four non-oiled sites in the Sound.

2. Determine the levels of hydrocarbons in tissues and eggs of brown king crabs at an oiled and unoiled site.

3. Determine the effects of exposure to petroleum hydrocarbons from the Exxon Valdez on both species of crabs through the assessment of 1) the reproductive condition of crabs in oiled and non-oiled sites by measuring such variables as fecundity, egg loss and larval production in crabs from these sites, 2) the incidence of limb loss and abnormalities in newly formed crab shells, and 3) the pathological effects, if any, of oil contamination on vital tissues and organs of crabs from oiled and unoiled sites.

4. Determine whether these observations demonstrate any adverse changes in viability.

5. Identify potential alternative methods and strategies for restoration of lost use, populations, or habitat where injury is identified.

METHODS

KING CRAB

This project will be conducted during September 1989. King crabs will be captured with commercial style crab pots (6' x 6'). All crabs captured will be utilized to fulfill either the hydrocarbon, necropsy, fecundity, injury or abundance estimate requirements of the project.

The Knight Island Passage area will be stratified by depth for pot sampling. Two strata (150 to 225 fathoms and ≥ 225 fathoms) will be defined within the Passage and each strata will be subdivided into sampling stations of approximately one square nautical mile. Sampling by strata will be employed to determine if hydrocarbon contamination has detectable differences with depth. If hydrocarbons are detected, necropsy samples will document the tissue and organ damage that has occurred.

Each station will be sampled with one pot for a total of 51 sampling locations. Additional pot sampling will be conducted in areas from Knight Island Passage where crabs appear abundant or oil is located by visual observation on the pots or crabs during the survey.

A total of ten pots will be set in Port Nellie Juan to obtain crabs for hydrocarbon, necropsy, injury and fecundity analysis as representatives of crabs from an oil free area.

A minimum of seven casts will be made with a SEA-BIRD water profiler to obtain data on temperature, salinity and dissolved oxygen.

If hydrocarbon uptake can be documented in brown king crab one manifestation of the contamination may be reduced egg production by

female crab. Pleopod samples from up to 50 ovigerous crabs will be collected and returned to Cordova for estimation of size-fecundity relationships. The collection of crabs and the fecundity size relationships will be repeated annually to estimate if a significant difference exists between years. Changes in fecundity data over time may be linked to oil contamination through necropsy of ovary and hydrocarbon analysis of tissue and egg samples.

Previous studies indicate that one effect of oil contaminated prey items is reduced growth (Gharrett et al. 1985, Karinen 1985). In an effort to monitor growth over time, spaghetti type tags will be applied to approximately one out of three crabs captured. This data can be compared to other tagging studies on brown king crab by the National Marine Fisheries Service from the Aleutian Islands. Increment of growth will be obtained from future surveys and through the commercial fishery.

DUNGENESS CRAB

This part of Fish/Shellfish Study Number 14 is a cooperative project between the National Marine Fisheries Service (NMFS), Auke Bay Laboratory and the Alaska Department of Fish and Game (ADF&G). The Alaska Department of Fish and Game will sample Orca Inlet in eastern Prince William Sound. The Orca Inlet portion of the project is an extension of an existing ADF&G project which began in July 1988 as part of the CFSO Shellfish Increment. The Auke Bay Laboratory will sample in pairs (impacted and non-impacted) 8 sites in western Prince William Sound to be selected from the following list: Sleepy, Iktua, Herring, Dryer, Paddy, Ewan, Main, and Eshamy Bays, Fox Farm and Snug Harbor.

Sampling will take place approximately every two months throughout the year (April, July, September and December) beginning in April 1989. The ADF&G field program will precede the sampling periods specified by the NMFS field program and will involve two to four days (1-3 days site work, 1 day travel) work during each sampling period throughout FY 1989 and 1990. The NMFS sampling will be conducted for two days at each site. Sampling periods will last 18 days (16 days site work, 2 days travel) every two months throughout FY 1989 and 1990.

Crabs in Orca Inlet will be sampled with Dungeness crab pots which will soak for 24 hours after which pots will be randomly selected for sampling. In western Prince William Sound NMFS will conduct systematic surveys using divers at depths to 30.5 meters (100 ft) during one or two days of each sampling period. Dungeness crab pots will be fished below 30.5 meters and will soak for 24 hours then randomly pulled for sampling.

In Orca Inlet three strings of 10 pots each will be set along the depth contour where crab are most abundant. Crab distribution will be isolated by setting strings of pots at multiple depths. Pot soak time will be decreased from 24 hours to 6 hours during the initial search phase. The depth of pot deployment may increase during fall and winter sampling periods as crab move deeper. In western Prince William Sound crabs will be sampled with Dungeness pot gear and divers. A ladder search will be conducted by two divers to estimate abundance of crabs to a depth of 30.5 meters. A string of ten pots will be set below 30.5 meters offshore of the diver zone. If crab are not found in the diver zone, then location of crab will be determined by repeated deployment of the string of 10 pots at multiple depths. Pots will soak for 6 hours during this search phase.

Diver observations will be recorded on number of crab and miscellaneous species seen, depth, slope, and substrate by distance traveled. Information on dive buddies, time underwater, visibility and direction of the ladder search will also be recorded. Information recorded during the pot sampling will include: time pot gear is set, pulled, and depth at which it is set, catch composition of each pot and sex, carapace width, fresh weight and external physical condition of all crab.

A total of 30 live female crab will be sampled from the site during each sampling period. Three samples of 10 crab each will be randomly selected from pot catches or from crab collected on a diver transect. In the pot sampling one sample will be formed by randomly selecting 1 female crab from the catch of each pot in a string of 10 pots. Therefore, 1 sample will be taken from each string of pots equalling 10 crab from each of the 3 strings of 10 pots. If no female crab occur in a pot, then another pot will be randomly picked and a female crab selected from it.

The specimen number, carapace width, fresh weight, shell age, egg color, clutch size, durometer reading and physical condition of each of the 30 crab will be recorded. In Orca Inlet three subsamples of three crab each will be randomly selected from the 10 crab in each sample to yield three composite hydrocarbon samples of ovaries and eggs. These nine crab will be measured and sacrificed for the hydrocarbon samples. In western Prince William Sound three crab will be randomly selected from the 10 crab in each sample to yield one composite hydrocarbon sample of ovaries and eggs. In all cases three ovaries will equal 1 composite hydrocarbon sample of ovaries. One composite hydrocarbon sample of eggs will be taken by clipping a small portion of the egg clutch (4 gm = 1/3 of pleopod) from the right fifth pleopod of each of the three crab in a subsample. The left fifth pleopod will then be removed from each of the ten crab to estimate egg development, egg mortality, egg fouling and infestation by predators. In Orca Inlet a single crab will remain from the sample of 10; in Western Prince William Sound 7 crab will remain. These crab will be returned to the sea.

During the second sampling period (July) an additional 7 crab from each string of pots will be sampled for necropsy. A number will be inscribed on the carapaces of the 7 crab in the sample. These crab will be placed in a flow-through tank until preparation for necropsy. Specific instructions for preparation of specimens for necropsy are in Appendix A.

Composite samples of ovaries will be placed in 16 oz glass jars rinsed with methylene chloride or heated to 440 degrees C for 4 hours. The jar will be labeled with site name, latitude, longitude, date, species, "ovary", sample number, specimen numbers of the sacrificed crabs, sampling team members, "DUNGENESS", and "ADF&G" or "NMFS". Composite samples of eggs will be placed in 16 oz glass jars rinsed with methylene chloride. The jar will be labeled with site name, latitude, longitude, date, species, "egg", sample number, specimen numbers of the sacrificed crabs, sampling team members, "DUNGENESS", and "ADF&G" or "NMFS". Jars containing samples will be frozen until hydrocarbon analysis is initiated. Pleopods will be placed in 18 oz whirlpacks or ziploc bags. Five percent neutral buffered formalin will be added to the bag to completely cover the pleopod. Each bag containing a pleopod will be labeled with site name, latitude, longitude, date, species, "pleopod", sample number, specimen numbers, sampling team members, "DUNGENESS", and "ADF&G" or "NMFS".

In eastern Prince William Sound sediment samples will be collected from Orca Inlet with a tom-tom corer during each sampling period. The tom-tom corer takes four core samples each time it is deployed. The corer will be deployed 3 times in Orca Inlet during each sampling period: once at a randomly selected point along each of the 3 strings of pot gear that are set along a single depth contour. Within a sampling period the corer will be deployed at the same depth each time. This will enable comparison of sediment samples with meiofauna samples which will be taken from the same depth within a site.

Core samples will be removed from the corer and placed in 16 oz glass jars that have been previously rinsed with methylene chloride or heated at 440 degrees C for 4 hours. Each jar will be labeled with site name, latitude, longitude, date, depth, "sediment", sample number, sampling team members, "DUNGENESS", and "ADF&G". Jars with sediment samples will be frozen at the lowest possible temperature until hydrocarbon analysis is initiated.

In western Prince William Sound sediment samples will be collected by divers in the subtidal zone and by a sampling team member in the intertidal zone. Eight samples will be taken by divers in the subtidal zone using hand operated syringe corers. Samples will be taken randomly along a transect traversing the area surveyed by divers. Each sample will be collected at the same depth. Intertidal samples will be collected from randomly selected points along a 30

meter transect run at the + 2.0 meter tidal height. Eight samples will be taken in the intertidal zone using hand operated syringe corers.

Sediment samples will be extruded from the syringe and placed in 16 oz glass jars that have been rinsed with methylene chloride or heated at 440 degrees C for 4 hours. Each jar will be labeled with site name, latitude, longitude, date, depth, "sediment", sample number, sampling team members, "DUNGENESS", and "NMFS". Jars with sediments will be frozen at the lowest possible temperature until hydrocarbon analysis is initiated.

Meiofauna samples will be collected only from the subtidal zone in eastern Prince William Sound. Samples will be collected with a tom-tom corer during each sampling period. The corer will be set next to the position where sediment core samples were taken. The corer will be deployed the same number of times and in the same manner for collection of meiofauna samples as it was for collection of sediment samples.

Meiofauna samples will be removed from the corer as follows: (1) a core cylinder will be carefully removed from the corer and any liquid remaining on the surface of the sediment will be poured into the sample container (4 oz jar); (2) the top 2-3 cm of sediment will be extruded from the cylinder by applying even pressure to the bottom of the sediment core; (3) the top 2-3 cm of sediment will be cut off the core over the sample jar into which it can fall undisturbed. Enough 10% formalin will be added to the jar to cover the sample. The jar will be labeled with the site name, latitude, longitude, date, depth, "meiofauna", sample number, sampling team members, "DUNGENESS", and "ADF&G".

In Western Prince William Sound meiofauna samples will be collected from intertidal and subtidal zones at each site. Samples will be collected by divers with a hand operated syringe corer. Samples will be taken next to the position where sediment core samples were taken at every site. The corer will be deployed the same number of times and in the same manner for collection of meiofauna samples as it was for collection of sediment samples.

The sample will be extruded from the syringe into a 4 oz jar. Enough 10 % formaldehyde will be added to the jar to cover the sample. Each meiofauna sample will be labeled with the site name, latitude, longitude, date, depth, "meiofauna", sample number, sampling team members, "DUNGENESS", and "NMFS".

Physical Oceanographic data will be collected at each site during each sampling period using an instrument that measures conductivity, temperature, and depth (CTD). Information will be collected and recorded by the CTD on depth, dissolved oxygen, temperature and conductivity of the water every 2 seconds as it is lowered to the bottom and raised to the surface. The CTD will be deployed 6 times

in Orca Inlet in eastern Prince William Sound during each sampling period. Measurements will be taken at each end of each string of 10 pots. In western Prince William Sound the CTD will be deployed 4 times at each site during each sampling period. Measurements will be taken at randomly selected points along a transect traversing the area surveyed by divers.

A total of 56 Dungeness crab will be tagged to study crab movement in western Prince William Sound. No crab will be tagged in eastern Prince William Sound. Seven female crab at each site in western Prince William Sound will be fitted with ultrasonic transmitters (Sonotronics) with a battery life of two years. Crab will be tagged during the first sampling period. The positions of these crab will be fixed during each sampling period using triangulation with a sextant used as a pelorus, and a portable loran.

In April 1990, 15 live ovigerous crabs will be collected from each site by divers and transported to the NMFS Auke Bay Laboratory. Specimen numbers will be inscribed on the carapace of each crab and specimen numbers for each site recorded. Depth of capture, carapace width, fresh weight and external physical condition of each crab will also be recorded. Crab will be held individually in flow-through tanks until larval release. Crab will be fed with a mixed diet of locally available prey including shrimp, clams and mussels. Water temperature, salinity and general condition of crabs will be regularly monitored. Timing of larval release, number of live and dead larvae, and larval swimming ability will be recorded to estimate larval production and viability.

Definitive analysis of the chemical composition of petroleum hydrocarbons in the sediments, tissues and eggs will be accomplished in the laboratory with gas chromatography/mass spectrometry at the Auke Bay Laboratory. Analyses will include 1) TPH/GC and PNA/SIM characterization of oil in marine sediments and crab tissues, 2) total organic carbon on selected samples, and 3) size fraction analysis on representative sediment samples. Prescreening analyses of collected samples will occur prior to full GC/MS analysis in areas of low liklihood of oiling. Details of the methods used in the chemical analyses are recorded under the Quality Assurance Program.

DATA ANALYSIS

KING CRAB

To address objectives one and two, statistics for analysis of variance will be computed to assess any differences in hydrocarbon content and incidence of tissue abnormalities between shallow verses deep strata in the oiled area of Knight Island Passage. Hydrocarbon and necropsy samples from Port Nellie Juan will be used as a

comparison for background levels from a suspected unoiled area.

To address objective 3 species data will be compiled to show detailed catches by pot. Cumulative data will be pooled and presented for all pots fished. Crab catches will be reported by size, sex, and relative age. Size frequency distributions will be presented by sex.

Fecundity data will be regressed to obtain a carapace length - egg count relationship. This line or lines of best fit will have an associated 95% confidence interval about the line.

Incidence of leg loss and other injuries will be recorded and summarized by depth.

To address objective four, data will be analyzed to determine level and cause of impact and suggest suitable mitigation and restoration techniques such as fishery restrictions and continued monitoring of stocks.

DUNGENESS CRAB

The number of paired sites with an adequate abundance of Dungeness crab that can be accessed within the time frame of this project is estimated to be between three and four. Four paired impacted and non-impacted sites provide four replicates for the two treatment levels. Three paired impacted and non-impacted sites would provide three replicates for the two treatment levels. Paired beaches were recommended for analysis of meiofauna data. Three to four replicates will provide large enough sample sizes within each impact level to minimize variance and enable detection of differences between impact levels if they exist. One control site is included from eastern Prince William Sound to insure data from a truly non-impacted area was collected in case non-impacted sites in western Prince William Sound are found to be contaminated. Should a priori categorization of sites fail, then 6 to 8 sites will be an adequate number to allow post stratification of sites to new treatment levels.

The number of specimens for one hydrocarbon analysis is dependent on the amount of tissue available in each crab and the need for a representative composite sample size. Ovarian tissue volume based on the average size of mature female crab was estimated and the number of specimens (3) required to provide 15 gm of tissue calculated. One pleopod could provide enough tissue for a 15 gm egg sample. However, a sample representative of more than one crab is desirable. Since three crab are needed for ovary tissues, the same three crab will have egg clips taken to form a composite egg sample for hydrocarbon analysis. Three hydrocarbon samples from each site are needed to detect contamination between impact levels (Dr. B. Clark, personal communication).

Eastern Prince William Sound crab hydrocarbon sample size was multiplied by the minimum number of replicates (3) in western Prince

William Sound because of the low number of samples (3) allocated for the only site in eastern Prince William Sound which comprises a complete impact level. Three replicates (sites) were used instead of 4 because: (1) consensus of biologists familiar with the Orca Inlet Dungeness crab stock is that a sample as large as 36 crab may not be available during all sampling periods; and (2) reducing the required harvest from 36 crab to 27 minimizes the overall impact of sampling on this uncontaminated stock of Dungeness crab.

A sample size of 30 crab is estimated as an adequate number to determine differences in reproductive capacity between impact levels based on data from Dungeness crab populations at log transfer facilities (LTF) in southeastern Alaska (O'Clair and Freese, 1988). Sample size for reproductive capacity is based on the difference between mean fecundity and mean worm infestation at 7 paired control sites and LTF sites, variance in fecundity and infestation by pair, probability of making a type I error equal to .05, and probability of making a type II error equal to .05 (Neter et al 1985, Table A-10). Data for mean fecundity and worm infestation were taken from O'Clair and Freese (1988). Sample size for detecting difference in mean fecundity and worm infestation between impact levels was estimated at 27 crab for fecundity and 26 crab for worm infestation. This sample was rounded up to 30 crab. The sample size for necropsy was determined by Dr. Ted Meyers, Alaska Department of Fish and Game.

A sample size of 8 was considered an adequate number of samples to detect a significant difference in sediment contamination or meiofauna abundance and diversity between tidal zones, sites, and impact levels (Dr. T. Shirley, personal communication). A slightly larger sample size of 12 will be taken in eastern Prince William Sound because: (1) a set of samples should be taken at each string of gear; and (2) larger samples sizes are desirable for the control level.

Data values for all physical oceanographic variables will be recorded every two seconds each time the CTD is deployed. Two deployments of the CTD at each site will probably be sufficient for summarizing the physical oceanography of each site because these parameters are fairly constant over short distances. Should a fresh water effluent occur near or at a site these parameters can change within a short distance. Therefore, during the first sampling period a larger number of casts systematically covering the sampling site is recommended. Subsequent comparison of data values from samples at each site may indicate fewer samples are needed during future sampling periods.

The number crabs chosen for ultrasonic tagging is based on: (1) work by O'Clair et al. (in press) in which 35 Dungeness crab were fitted with ultrasonic tags in five bays in southeastern Alaska and tracked for periods of about a week; and (2) the number of ultrasonic tags with unique pulse codes currently available.

Ten crab from each site is estimated to be an adequate number to determine differences in larval production between sites based on laboratory studies done on Dungeness crab and other species at the Auke Bay Laboratory (C. O'Clair, personal observations). Fifteen crab will be collected from each site to insure that an adequate number of crab survive transportation to holding facilities.

All data will be tested for heteroscedasticity with Bartlett's test or equivalent (Sokal and Rohlf 1981). Parametric statistics (analysis of variance and Scheffe's a posteriori test) will be used to test for differences in means between oiled and non-oiled sites if underlying assumptions of the parametric procedures are met, otherwise nonparametric tests (eg. the Kruskal-Wallis test) will be employed. For the purposes of the following discussion we assume that the data will meet or can be transformed to meet the assumptions of the parametric tests.

Objective 1 will be addressed by a multi-part analysis. First paired t-test statistics (or two-way ANOVA) for hydrocarbon content in crab and sediment will be calculated for paired sites at each area. This will determine if paired oiled and non-oiled sites have been categorized correctly according to level of contamination. Results of these tests may require post stratification of sample sites into categories other than oiled, non-oiled and control. Second, analysis of variance will be used to test for differences in hydrocarbon content in Dungeness crab tissues among areas and between oiled, non-oiled and control sites. Third, analysis of variance will be used to test for differences in hydrocarbon content in sediments between oiled, non- oiled and control sites. Comparisons with the control site will indicate the severity and extent of impact by area.

Under objective 3 analysis of variance will be used to test for differences in reproductive parameters (fecundity, egg mortality, egg fouling and infestation rates of egg predators) among areas and between levels of impact. Correlation analysis will be used to relate to hydrocarbon levels of sediments to reproductive parameters. Analysis of variance can also be applied in the above manner to detect differences in: (1) results of histological analyses of crabs collected at each site; and (2) larval production and viability of crabs collected and held until larval release. These two analyses will provide additional information for evaluating objective 3. Graphic analysis of direction and distance traveled by crabs tagged with ultrasonic transmitters in oiled and non-oiled areas can be compared to detect avoidance of contaminated areas. Differences in the pathological effects of oil contamination on crabs between levels of impact will be addressed with analysis of variance. These differences can then be related to hydrocarbon concentratiions in the corresponding crab tissues and sediment.

The abundance and diversity of meiofauna among areas and between levels of impact will be subjected to analysis of variance. The results will be related to levels of contamination in sediments with correlation analysis.

Further multivariate statistics (eg. analysis of covariance, rank correlation coefficients, discriminant analysis) will be computed if the above summary statistics indicate relationships may exist between Dungeness crab hydrocarbon content, histological condition, reproductive capacity, sediment hydrocarbon content, meiofauna abundance and diversity, and physical oceanography factors.

SCHEDULES AND REPORTS

Date(s)	Activity
April - December 1989	Dungeness field collections
August 31 - September 7, 1989	King crab field collections
October - November 1989	Data entry, analysis and laboratory work
December 21, 1989	Preliminary report on impacts of oil on crabs
February, 1990	Final report on impacts of oil on crabs

PROJECT BUDGET [1]

Line Item	Category	Budget		
		ADF&G	NMFS	Totals
100	Personnel Services	20,600	30,000	50,600
200	Travel	1,500	4,000	5,500
300	Contractual	38,000	20,000	58,000
400	Commodities	4,800	2,000	6,800
500	Equipment	0	22,000	22,000
700	Grants	0	0	0
Total		64,900	78,000	142,900

[1] Budget is for all activities performed from March 27, 1989 to

February 28, 1990.

FUNDED PERSONNEL

Class	PCN	Name	PFT_mm	SFT_mm	Cost
FB I	11-1649	Trowbridge	4.0		18.5
FT II	N-217	Wright		2.0	4.5
GS-12		O'Clair	3.5		22.6
GS-9		Freese		2.0	8.4

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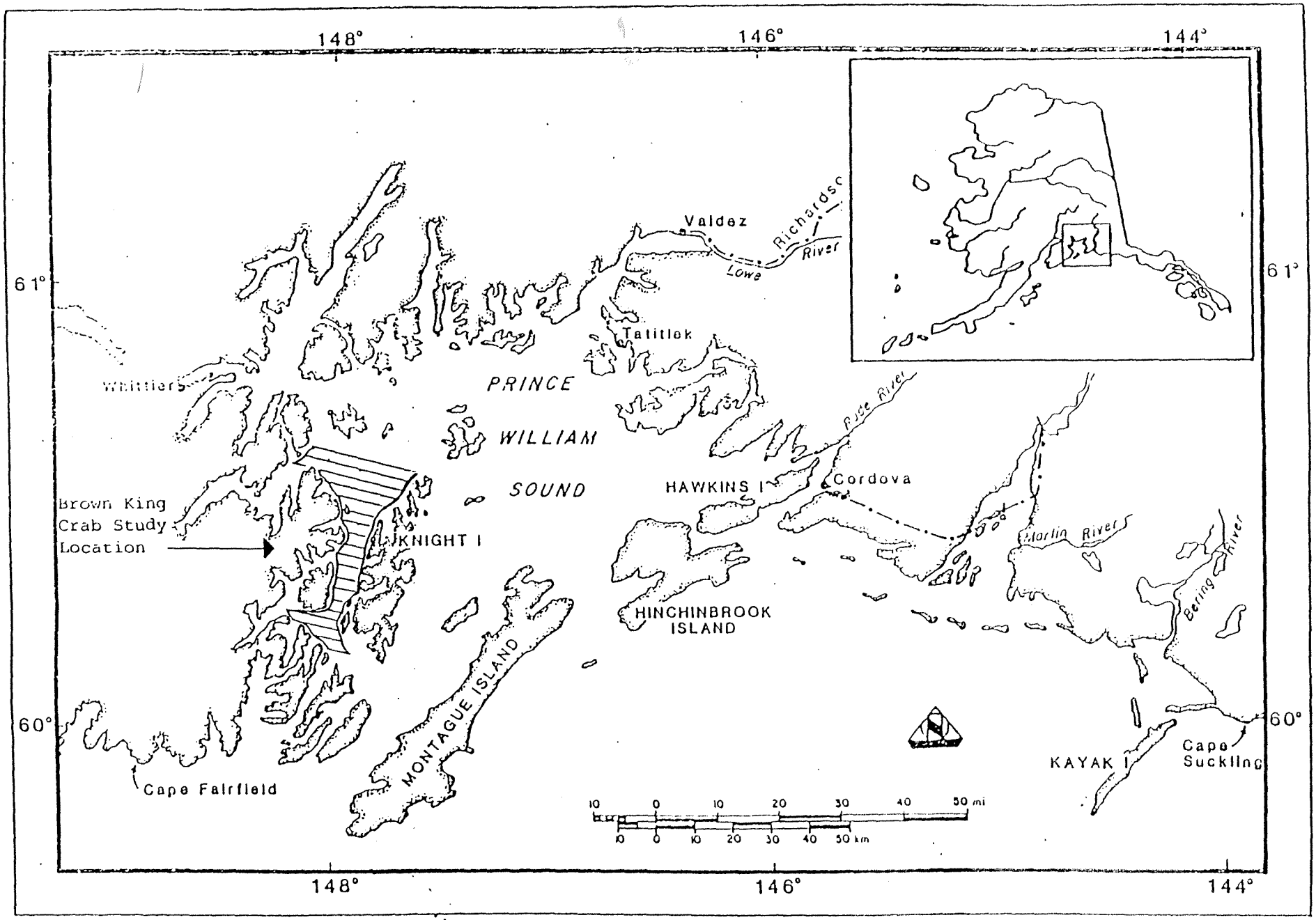
148°
146°
144°
61°
60°
Valdez
Richardson
River
Lowe
Tatitlek
Whittier
PRINCE
WILLIAM
SOUND
Brown King
Crab Study
Location
KNIGHT I
HAWKINS I
Cordova
Ridge River
Copper River
Bering River
HINCHINBROOK
ISLAND
MONTAGUE ISLAND
KAYAK I
Cape
Suckling
Cape Fairfield
10 0 10 20 30 40 50 mi
10 0 10 20 30 40 50 km

ADF&G BROWN KING OIL PROJECT SITE DESCRIPTION DATA FORM

Date _____ Skipper Name _____

Vessel Name _____ Vessel Ak Number _____

Vessel Description _____

Sampling Team	Agency

	Code	Description
Waves		
Weather		

Air Temp _____ Sea Temp _____

Wind Speed _____ Wind Direction _____

Buoy	Station #	Time Set	Time Pull	Soak	Depth	Latitude	Longitude

Waves

Code	Description
1	Glassy
2	Rippled
3	Wavelets
4	Slight 2-4'
5	Moderate 4-8'
6	Rough 8-13'
7	Very Rough 13-20'

Weather

Code	Description
1	Clear
2	Partly Cloudy
3	Overcast
4	Fog or Thick Haze
5	Showers
6	Squalls
7	Drizzle
8	Rain
9	Rain and Snow
10	Snow
11	Blizzard

ADF&G BROWN KING CRAB SAMPLING FORM

Station #_____ Date_____
Pot Id #_____ Sampler_____

FISH _____

SPEC IMEN	USE	SPEC IES	C.L.	LEGAL Y/N	C.W.	SHELL COND	EGG COND	EGG COLO	% FULL	PARA SITE	TAG #	SEX	INJU RIES

Species

1=Brown King
2=Red King
3=Blue King
4=C.bairdi
5=Dungeness

Shell Condition

0=soft
1=new
2=old
3=v.old

Egg Color

0=no eggs
1=purple
2=brown
3=orange
4=purple-brn
5=pink

Egg Condition

0=immature
1=not eyed
2=eyed (new)
3=eyed (old)
4=eyed eggs & empty cases
5=empty cases
6=dead eggs

Use

1=Hydro
2=Nec
3=Both
4=Tag
5=Eggs
6=Return

Parasite

0=none
1=rhizocephalen
2=blk-mat (L)
3=blk-mat (M)
4=blk-mat (H)

Clutch size

0=immature
1=mature, no eggs
2=trace
3=1/8 full
4=1/2 full
5=full
6=eggs bulging

Sex

1=male
2=female

ADF&G BROWN KING CRAB
OIL SPILL ASSESSMENT
NECROPSY SAMPLE FORM

	CRAB 1	CRAB 2	CRAB 3	CRAB 4	CRAB 5
POT ID.#					
SPEC. #					
TISSUES					
Blood smear					
Carapace					
Heart					
Rt. gill arch					
Epidermis					
Hepatopanc.					
Green glands					
Gonads (3pcs.)					
Esophagus					
Cardiac stomach					
Pyloric stomach					
Midgut					
Hindgut					
Rectum					
Thoracic gang.					
Cerebral gang.					
Eyestalks					

ADF&G BROWN KING CRAB SAMPLING FORM

Station #_____ Date_____
Pot Id #_____ Sampler_____

FISH_____

SPEC IMEN	USE	SPEC IES	C.L.	LEGAL Y/N	C.W.	SHELL COND	EGG COND	EGG COLO	% FULL	PARA SITE	TAG #	SEX	INJU RIES

Species

1=Brown King
2=Red King
3=Blue King
4=C.bairdi
5=Dungeness

Parasite

0=none
1=rhizocephalen
2=blk-mat (L)
3=blk-mat (M)
4=blk-mat (H)

Shell Condition

0=soft
1=new
2=old
3=v.old

Clutch size

0=immature
1=mature, no eggs
2=trace
3=1/8 full

Egg Color

0=no eggs
1=purple
2=brown
3=orange
4=purple-brn
5=pink

4=1/2 full
5=full
6=eggs bulging

Egg Condition

0=immature
1=not eyed
2=eyed (new)
3=eyed (old)
4=eyed eggs & empty cases
5=empty cases
6=dead eggs

Sex

1=male
2=female

Use

1=Hydro
2=Nec
3=Both
4=Tag
5=Eggs
6=Return

ADF&G BROWN KING CRAB
OIL SPILL ASSESSMENT
NECROPSY SAMPLE FORM

	CRAB 1	CRAB 2	CRAB 3	CRAB 4	CRAB 5
POT ID.#					
SPEC. #					
<u>TISSUES</u>					
Blood smear					
Carapace					
Heart					
Rt. gill arch					
Epidermis					
Hepatopanc.					
Green glands					
Gonads (3pcs.)					
Esophagus					
Cardiac stomach					
Pyloric stomach					
Midgut					
Hindgut					
Rectum					
Thoracic gang.					
Cerebral gang.					
Eyestalks					

ADF&G BROWN KING CRAB
OIL SPILL ASSESSMENT
HYDROCARBON SAMPLE FORM

POT ID.#	SPEC.#	HYDROCARBON EGGS	TISS	PANC.	HYDROCARBON SAMPLE #	DATE

ADF&G EXXON VALDEZ OIL SPILL IMPACT ASSESSMENT TRIP REPORT FORM

Project

Dates ____ through ____

Field trip leader ____ Signature ____ Date

Purpose of trip ____

Types of Data Collected (physical, biological, & non-hydrocarbon samples)

Data	in Notebook #(s)

Physical Location of Notebooks:

Physical Location of Backup Copies:

Notes, comments:

Summary of hydrocarbon-analysis samples taken

Sample types	No. of samples	Chain of custody form #
Water		
Sediment		
Biological; species, tissue, whatever		

Notes, comments:

Reports of Oil Spill Impact Assessment Data

Report Name and Number:

September 12, 1989

STATE/FEDERAL DAMAGE ASSESSMENT PLAN

ANALYTICAL CHEMISTRY

COLLECTION AND HANDLING OF SAMPLES

FOR AGENCY USE ONLY
NOT FOR RELEASE
ATTORNEY WORK PRODUCT

TABLE OF CONTENTS

1. Introduction

In response to the release of more than 10 million gallons of crude oil into Prince William Sound, the State of Alaska and four Federal Agencies, the Departments of Agriculture, Commerce and Interior and the Environmental Protection Agency are acting together to assess the damages to the natural resources. Authority for this action is provided by the Comprehensive Environmental Response, Compensation, and Liability Act (CERCLA) and the Clean Water Act (CWA).

A damage assessment requires documentation of the exposure of the resources to oil released from the EXXON VALDEZ, identifying which resources were injured by that exposure, measuring the magnitude of the adverse affects on each resource over time and assigning economic values for that injury. Once this is done, monetary compensation can be sought from the potentially responsible parties to restore and/or replace the injured resources.

Recovery of monetary damages may involve civil court actions. It will then be necessary to prove that the samples were collected in a scientifically approved manner and that the samples were protected from outside contamination (non-incident related) and accidental mix-ups during handling and analyses. It is, therefore, extremely important that every sample be readily identified and their location and analytical status known and documented at all times.

This document and the associated training sessions, were prepared to assist field personnel in collecting samples that will provide scientifically sound and legally defensible data to support the State/Federal Natural Resource Damage Assessment for the EXXON VALDEZ oil spill.

2. Record Keeping and Documentation

Standard operating procedures (SOPs) for all sampling procedures, including chain of custody procedures; sampling protocols; cleaning and preparation of sample collection and storage devices; and labeling, handling, and sample

preservation and holding time must be written in detailed, clear, simple and easy to follow language.

Personnel must be knowledgeable and experienced in the described sampling techniques and must adhere to the SOPs.

Any changes in procedures must be recorded in detail in the field logbook. The log entry must include reasons that the change in procedure was unavoidable.

Field logbooks are issued by the Team Leader or their representative. The logbooks should be serially numbered, sturdy, bound books with sequentially numbered pages. Waterproof logbooks should be used if available.

Field data sheets, if used, must be consecutively numbered by project. The field data sheets must be referred to in entries in logbooks which reference, the precise data sheet involved and the relationship to specific data in the logbook noted.

All information pertinent to field activities, including descriptive notes on each situation, must be recorded in indelible marker in the field logbook. The information must be accurate, objective, up-to-date and legible. It should be detailed enough to allow anyone reading the entries to reconstruct the sampling situation. Additional information may be provided by field data sheets, sample tags or photographs.

Entries should be made in the logbook or on field data sheets with indelible marker at the earliest possible time. Notes should never be written on scrap paper and then transferred to the logbook.

Entries into field logbooks or field data sheets are signed or initialed, and dated by the person making the entry at the time of entry.

Each day's entries are closed out with a horizontal line, date and initial.

Errors in field logbooks or other records are corrected by drawing a single line though the error, entering the correct information and signing and dating the correction. Never erase an entry or any part of an entry.

Do not remove pages from the logbook.

Completed logbooks and field data sheets are returned to the Team Leader or their representative to be archived in a central location under chain-of-custody procedures until the Trustees indicate that they may be released.

3. Sample Identification and Labelling

A tag or label identifying the sample must be completed and attached to each sample. Waterproof (indelible) marker must be used on the tag or label. The minimum information to be included on the tag are the sample identification number, the location of the collection site, the date of collection and signature of the collector (who, what, where & when). This information and any other pertinent data such as the common and scientific names of the organism collected, the tissue collected and any remarks are recorded in the logbook. Field sample data sheets, photographs, any pertinent in-situ measurements (such as temperature, salinity, depth) and field observations are recorded in the logbook.

The location of the sampling site is determined with the aid of USGS grid maps, NOAA charts or navigational systems such as LORAN C. The site locations should be plotted on a chart of appropriate scale and photocopies incorporated into the logbook. In addition, a clear, detailed descriptive location as well

as the latitude and longitude, in degrees, minutes and seconds, of the collection site must be recorded in the logbook.

4. Sampling Equipment and Sample Containers

All sample containers must be either organic-free (solvent-rinsed) glass or organic-free (solvent-rinsed) aluminum foil. Lids for the glass containers must be lined with either teflon or solvent-rinsed aluminum foil.

Certified-clean glass jars are available from various vendors and if obtainable, may be used without cleaning.

Sample collection and storage devices are cleaned by washing with soap and hot water, rinsed extensively with clean water and then rinsed with either methylene chloride or acetone followed by pentane or hexane and allowed to dry before use.

First rinse: tap water, then re-rinse in distilled water.

Second rinse: methylene chloride or acetone

Third rinse (if acetone is used): pentane or hexane

The solvents (methylene chloride, acetone, pentane and hexane) used for cleaning sample collection and storage devices must be of appropriate quality for trace organic residue analysis and be stored in glass or Teflon containers, not plastic.

New glass jars or unused aluminum foil do no need to be washed with soap and

water. They must however, be solvent-rinsed as described above before use.

Glass jars may be cleaned by heating to 440°C for a minimum of 1 hour.

Clean glassware should be stored inverted or tightly capped with either solvent-rinsed aluminum foil or teflon-lined caps.

The dull side of the aluminum foil should be the side that is solvent-rinsed. Pre-cleaned squares may be stored with the clean sides folded together.

All equipment that comes in contact with the sample such as dredges or dissecting equipment must be solvent-rinsed before contacting each sample. Equipment should be steam-cleaned or washed with soap and hot water at the end of each day or between sampling locations.

5. Sampling Procedures

The method of collection must not contaminate the samples. Do not collect any subsurface samples through surface slicks. Do not collect any samples with oil-fouled equipment, such as nets or dredges. Do not touch or collect any sample with your bare hands.

Sample container volume must be appropriate to sample size; fill the jar to just below the shoulder. Overfilled jars will break when they freeze; underfilled jars will allow the sample to dry out.

At least one field blank and replicate sample should be taken for each collection site, batch of samples or 20 samples taken. (A field blank is a sample container opened in the field, closed and stored as if it contained a sample. A replicate sample is a second sample from the same site.) Rinsate blanks should be taken if appropriate.

5.1 Water - The method must be described or adequately referenced in sampling SOPs. Recommended sample size is 1-4 litsrs depending on the analytical methodology.

Water samples for volatiles analyses should be taken in 40 ml amber vials with no head space or bubbles.

5.2 Sediment - Any accepted methods of collecting undisturbed surface sediment samples such as box cores, hand corers, or grabs may be used. The method must be described or adequately referenced in sampling SOPs. Recommended sample size is 10-100 grams (a 4 oz. jar).

5.3 Tissue - Organisms to be analyzed for petroleum hydrocarbons should be freshly killed or recently dead. Decomposed organisms are rarely of any value for analysis.

Whole organisms may be stored in solvent-rinsed glass jars or wrapped in solvent-rinsed aluminum foil.

Tissue sections may be taken either on site from freshly killed organisms or in the laboratory from carefully collected and preserved - cold or frozen - whole organisms. Tissue should include flesh and internal organs, especially liver. Recommended sample size is 10-15 grams.

Tissue samples need to be protected from external contamination at time of collection. Contents of the intestinal tract, external slime coating, contaminated collecting utensils, etc. are all potential sources of contamination when collecting internal tissue samples.

All instruments used in handling samples must be made of a non-contaminating material (e.g. stainless steel, glass, teflon, aluminum) and solvent-rinsed between each sample collection.

Instruments used for exterior dissection must not be used for internal dissection.

Avoid hand contact with tissue sample.

Collect stomach and intestinal tract last.

Bird eggs are wrapped in solvent-rinsed aluminum foil and transported by any convenient means that will prevent breakage. They should be opened or refrigerated as soon as possible. Eggs are opened by cutting them with a solvent-rinsed scalpel or by piercing the air cell end and pouring/pulling the contents out. Avoid including pieces of egg shell with the contents or touch-

ing the contents with your hands. Total weight, volume (measured or calculated), length, width and contents weight must be recorded for each egg. Bile is collected by removing the gall bladder, puncturing it with a scalpel fitted with a new #11 blade, and collecting the contents in a 4 mL amber glass vial.

6. Sample Preservation and Holding Time

Samples must be kept cool, i.e. on ice.

Samples that are to be frozen, sediment and tissue, should be frozen quickly and rapidly. That is, these samples should be frozen as soon after collection as possible and the freezing process should be rapid.

Frozen samples must be kept frozen, at -20°C or less, until extracted or prepared for analysis. Repeated freezing and thawing of samples can destroy the integrity of the samples resulting in questionable data or the loss of data.

6.1 Water - All water samples must be immediately extracted with methylene chloride or preserved with HCl to pH<2. If preserved, water samples are stored in the dark at 4°C and extracted within 7 days. All extracts must be stored in the dark in air tight chemically clean containers until analysis.

6.2 Sediment and Tissue - Samples should not be extracted until immediately before analysis; if there is a lag between sample extraction and sample analysis, extracts must be stored in air tight containers kept in the dark at 4°C.

7. Sample Shipping

All samples, except water samples, must be kept frozen throughout the shipping process.

Samples must be packaged to prevent breakage. Glass jars should be individually wrapped so that they will not contact each other if padding shifts in transit (which styrofoam chips do). Bubble wrap or the divided boxes that new jars are shipped in work well. Pack samples in insulated containers (e.g. ice chests) with enough frozen mass to remain frozen in transit.

It is the responsibility of the sample shipper to arrange for sample receipt. Do not send samples off without arranging for pickup and storage.

To insure that samples are not compromised, shipment should not be initiated later in the week than Wednesday nor should samples be shipped in any week in which there is a holiday.

Shipments must comply with Department of Transportation regulations.

8. Chain-of-Custody Procedure

Samples must be kept in such a manner that they cannot be altered either deliberately or accidentally. Any indication that a sample has been subjected to tampering or physical alteration could disqualify it as evidence for possible

legal action.

The field sampler is personally responsible for the care and custody of the samples collected until they are transferred under chain-of-custody procedures.

A sample is considered in "custody" if:

it is in your actual physical possession or view;

it is retained in a secured place (under lock) with restricted access

or it is placed in a container and secured with an official seal(s) such that the sample cannot be reached without breaking the seal(s)

Evidence tape or sample seals are used to detect unauthorized tampering of samples following sample collection. The seal must be attached in such a way that it is necessary to break it in order to open the container. Seals must be affixed to the container before the samples leave the custody of sampling personnel.

All samples must be accompanied by a chain-of-custody record or field sample data record (Figure 1). When samples are transferred from one individual's custody to another's, the individuals relinquishing and receiving the samples will sign and date the chain of custody record. This record documents the transfer of custody of samples from the sampler to another person or to a specified analytical laboratory.

Shipping containers must be custody-sealed for shipment. The seal must be signed before the container is shipped. The chain-of-custody record must be dated and

signed to indicate any transfer of the samples. The original chain-of-custody record accompanies the shipment; a copy is retained by the sample shipper. If samples are sent by common carrier, copies of all bills of lading or air bills must be retained as part of the permanent documentation.

Whenever samples are split, a separate chain-of-custody record is prepared for those samples and marked to indicate with whom the samples are being split.

N.M.F.S. Prince William Sound Oil Assessment Chain of Custody Form

VAL-89- Project _____ Serial #

Page _____ of _____

NOTE: Use ballpoint pen, waterproof ink (eg Rapidograph) or fine-tip waterproof marker

NMFS Auke Bay Fisheries Laboratory
Box 210155, Auke Bay AK 99821
For information contact Sid Korn
(907) 789-6021 or 789-6000

Date Collected	Sample # (collector's)	Assigned # (leave blank)	Type (tissue, water, sediment, etc)	Location Collected	Latitude	Longitude	Remarks
5							
10							
15							
20							

Continue on back of page

Chain of Custody

Samples collected by _____ (print name) of _____ (agency) _____ (signature) _____ (date)

Relinquished by _____ (signature) _____ (date) at _____ (place) _____ (comments)

Recieved by _____ (print name) of _____ (agency) at _____ (place) _____ (signature) _____ (date)

Relinquished by _____ (signature) _____ (date) at _____ (place) _____ (comments)

Recieved by _____ (print name) of _____ (agency) at _____ (place) _____ (signature) _____ (date)

Relinquished by _____ (signature) _____ (date) at _____ (place) _____ (comments)

Recieved by _____ of _____ at _____ _____ (signature) _____ (date)

HISTOPATHOLOGY TECHNICAL GROUP

Justification/Concern

Histopathology is an important tool used in determining mechanisms of death and sublethal effects caused by infectious agents and toxic substances. A definitive diagnosis often does not result from histological examination, but can give strong support to other positive measurements. Tissues deteriorate (autolyze) rapidly after an animal dies; therefore, to be of value, any sample taken for histological evaluation as part of the damage assessment of the Exxon Valdez oil spill must be collected, preserved, and processed under strict guidelines.

Introduction

This committee was established to serve as an ad-hoc advisory and technical control group that reports to the Management Team. Its specific function is to serve as a control point for all laboratory aspects of histopathological analysis associated with the Exxon Valdez oil spill assessment program. This includes the development of detailed sampling protocols, appropriate training of field personnel in collecting samples, review of all histological sampling proposed and identification of effort duplication, establishment of a secured repository for all histology samples for storage until processing, oversee archiving and inventory of collected samples, qualification evaluation of potential subcontractors to be hired for processing and interpretation of histology samples, quality control assurance in all work performed, advice on chain-of-custody guidelines, and development of budget estimates to accommodate the required histopathological analyses.

TABLE OF CONTENTS - Histopathology Technical Group

1. Sample Collection and Preservation Protocols

Standard protocols for necropsy and preservation of tissue samples (including a materials list and catalog numbers) for histopathology described in the appendices shall be used throughout the oil spill assessment studies. Different protocols have been designed to accommodate the different groups of animals to be encountered in the assessment studies. Necropsy procedures are included for:

Appendix 1: Finfish
Appendix 2: Bivalve molluscs
Appendix 3: Brachyuran and crab-like Anomurans; i.e., King crabs)
Appendix 4: Shrimp
Appendix 5: Marine and terrestrial mammals
Appendix 6: Migratory and nonmigratory waterfowl

Paired sampling of animals from oiled versus non-oiled sites will be done for comparative purposes. Histopathological sampling should be done during any observed acute episodes of mortality or morbidity to determine the cause of death or abnormality. These types of samples are the most valuable in assessing acute toxicity affects and will be the most likely samples collected for birds and mammals due to their high visibility in the impacted areas. Because of the low visibility of fish and shellfish, many histology samples will consist of random collections in impacted and control areas with little prior obvious indication of morbidity or mortality.

Any histological processing of samples collected from apparently normal shellfish should be performed after results of parallel hydrocarbon sampling is known; i.e., positive hydrocarbon results may merit further histopathology studies. This would not be advisable for fish and other higher animals that possess an active mixed function oxidase (MFO) liver enzyme system which could metabolize hydrocarbons to other compounds providing negative hydrocarbon results, but potential toxicological lesions. Other enzyme function analyses being performed, such as on bile, may show an activated MFO system in exposed fish and higher animals. Consequently, histology and hydrocarbon samples, as well as other appropriate samples, such as bile, for metabolite and enzyme function analyses, should be taken from the same animal when possible. If certain fish and shellfish are too few or small, subsampling other animals from the same site at the same time will be necessary.

2. Processing and Interpretation Protocols

Histopathology assessment of birds and mammals will be done primarily on tissues from clinically affected animals using established criteria of cellular degenerative and necrotic changes recognized by any board-certified veterinary pathologist.

Histopathological analysis of finfish and shellfish tissues will include the criteria above as well as indices established in the Amoco Cadiz oil spill studies (Haensly et al. 1982; Berthou et al. 1987) to allow some quantification of potentially subtle degenerative changes in tissue histology of otherwise clinically normal animals. Briefly these indices include:

a. Mean concentration of mucus cells per mm^2 of gill lamellae (fish).
b. Mean concentration of mucus cells per mm of epidermis in 10 fields (fish).
c. Average epidermal thickness in mm measured in 10 fields (fish).
d. Mean concentration of macrophage centers per mm of liver.

e. Mean concentration of hepatocellular vacuolation due to fatty degeneration (fish).
f. A mean and total tissue necrosis index (invertebrates).
g. Histological gonadal index (invertebrates).
h. Differences in prevalences and intensities of incidental lesions caused by infectious agents (fish and invertebrates).

3. Quality Assurance in Field Collection of Samples and in Interpretation of Results

Field Collection

Veterinary personnel trained in sample taking should be utilized for on-site necropsies of birds and mammals in order to ensure adequate quality control and standardized sample collection in these less familiar and more complex species. The same high standards should be attainable in fish and invertebrates if sample collection is done by trained finfish and shellfish biologists. A fish pathologist and technician will be available to train field personnel and assist in necropsy and preservation of finfish and shellfish samples at collection sites.

Sample collection from migratory birds and sea otters should be coordinated with the U.S. Fish and Wildlife Service National Wildlife Health Laboratory in Madison, Wisconsin. Collection of samples from nonmigratory birds and other marine mammals could be coordinated with the Alaska State Veterinary Laboratory in Anchorage. Finfish and shellfish samples can be coordinated through the on-site fish pathologist and the ADF&G, Fisheries Rehabilitation, Enhancement and Development (FRED) Division Juneau Fish Pathology Laboratory.

Interpretation of Results

Quality control of all processed work will require independent blind reading of subsampled histology slides by two different laboratories.

Tissues with known lesions will be included periodically in groups of tissue samples for blind reading and determination of competency in interpretation.

4. Repository For Samples And Inventory Procedures

A common repository for storage of all histology samples awaiting processing will be established at Anchorage in a secured building in compliance with chain-of-custody requirements. Samples received will be given a unique accession number to be cross-referenced with the project and original numbering assigned by the collector.

5. Chain-Of-Custody Guidelines

Due to the evidentiary nature of sample collecting investigations, the possession of samples must be traceable from the time the samples are collected until they are introduced as evidence in legal proceedings. To maintain and document sample possession, chain-of-custody procedures must be followed.

The field sampler will be personally responsible for the care and custody of the samples collected until they are transferred. All samples will be accompanied by a chain-of-custody record or field sample data record (Appendix 7). When samples are transferred from one individual's custody to another's, the individuals relinquishing and receiving will sign, date, and note the time on the record. This record documents the transfer of custody of samples from the sampler to another person and, ultimately, to a specified analytical laboratory.

Shipping containers will be custody-sealed for shipment. This procedure includes use of a custody seal such that the only access to the package is breaking the seal. The seal shall be signed before the sample is shipped. The chain-of-custody record will be dated and signed to indicate transfer. The original record will accompany the shipment and a copy will be retained by the sample collector. Whenever samples are split, a separate chain-of-custody record will be prepared for those samples and marked to indicate with whom the samples are being split.

If samples are being sent by common carrier, copies of all bills of lading or air bills must be retained as part of the permanent documentation.

6. Subcontracting for Histological Work

Subcontracting work for histopathology processing and interpretation should be coordinated through the Histology Technical Group which will determine if selected processors are qualified to do the work. Qualifications for mammal and avian samples will require a board-certified veterinary pathologist. Finfish and shellfish work will require individuals with a demonstrated publication record in the field of histopathology.

7. Finfish and Shellfish Mortality Assessments

Estimates of finfish and shellfish mortalities will be according to guidelines established for estimating fish kills contained in Part II (Fish Kill Counting Guidelines) of the Monetary Values of Freshwater Fish and Fish-Kill Counting Guidelines, American Fisheries Society Special Publication Number 13, 1982, including use of appropriate random sampling methods and tagged carcasses (Natural Resource Damage Assessments provided by CERCLA).

8. References

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9. Appendices (attached)

HISTOLOGICAL SAMPLE PREPARATION FOR BRACHYURAN AND ANOMURAY CRAB SPECIES

Histopathological Technical Group

NOTE: Only live or moribund crabs will be suitable for processing. Histopathological changes caused by toxic chemicals are often very subtle at best. Tissues in dead crabs autolyze very quickly and will mask these changes. Do not collect and process dead crabs. Keep crabs alive in containers of seawater or live wells if they must be transported to the processing site. Do not over-ice animals such that tissues freeze while in transit. Frozen tissues are worthless for histological examination.

1. The fixative to be used is 10% neutral buffered formalin solution (formula attached). Formalin should be handled wearing rubber or latex gloves.

2. The volume of fixative should be ten times the volume of the tissue. This is important since any less fixative may result in tissue autolysis and worthless samples. After 72 hours, the formalin should be poured off and replaced with 70% ethyl alcohol for storage and transport. This accomplishes an important objective; i.e., it prevents tissues from becoming too hard and brittle when stored in fixative for long periods. Also, the fixative poured off may be saved and strained of tissue fragments and used one more time for preserving other samples.

3. The sample size per site or species will be 20 crabs, live or moribund.

4. Prior to tissue collection, a blood smear should be prepared from each live crab. Insert a 1-cc syringe with a 20-gauge needle into the articular membrane of any walking leg. The third joint of either cheliped works best. Express a large drop of blood from the syringe onto one end of a clean, frosted-end glass slide and use another slide to make the smear as illustrated in the attached information. Allow to air dry, label the frosted end with an assigned crab number, and date and include in a small slide box with the samples below. An alternative method would be to pull off a walking leg and allow not more than 1-2 drops of blood to fall onto the slide.

   Be sure to not let salt water mix with the blood on the slide, as it will cause blood cell lysis. Note: King crab blood clots unbelievably fast, so make your smear quickly.

5. The chitinous exoskeleton of large crustacea prevents adequate penetration of any fixative by simple immersion. Consequently, major organs and tissues of crabs must be dissected out and dropped into fixative. This procedure is described by the following:

   a. The carapace over the visceral cavity of the crab must be removed using tin snips or bone snips, or otherwise heavy duty serrated scissors (Figure 2).

   b. Once the carapace is removed, the pigmented epidermis may come off attached or remain overlying the viscera. Snip a small 5-mm portion of the

epidermis overlying the heart and save for fixation, then proceed to uncover the visceral cavity. Also, fix a 1-cm square piece of the carapace.

c. Once the cavity is exposed, the heart, cardiac stomach, hepatopancreas, gonads (posterior to heart in Alaskan crabs), and gills become obvious (Figure 4).

d. Remove the right rear gill arch and take a 0.5-cm portion thereof.

e. Remove a 0.5-cm portion of the heart which will be beating if the animal has been freshly killed.

f. Remove a 0.5-cm square of hepatopancreas to the left of the heart.

g. Remove both antennal glands (green glands).. Each lies on either side against the frontal carapace of the crab and is surrounded by urinary bladder and hepatopancreas (Figure 7). This can be a difficult organ to find and should be retrieved early on before other tissues are disturbed and landmarks are lost.

h. Remove the entire GI tract starting with the esophagus, which is ventral and anterior to the cardiac stomach (Figure 4) continuing with the entire stomach and intestine ending with the rectum that terminates at the vent on the ventral surface of the abdominal apron or flap underneath the crab. The intestine is long, curling down posterior to the heart (Figure 4) and extending anteriorly into the abdominal flap. It is fragile and requires some digging with forceps and cutting away from hepatopancreas with scissors to free the specimen. Remove 0.5-cm portions of the esophagus, cardiac stomach, pyloric stomach, midgut, hindgut, and rectum.

i. Remove a 0.5-cm section of the gonads also located posterior to the heart on either side. Gonads are part of the tissue in the way of extracting the intestine.

> Ovaries are large diameter, tubular organs that can be white, yellow, blue, or dark brown in color, depending upon the crab species.
>
> Testes are thin, very white, twisted threads containing viscous gametogenic material. Remove anterior, mid, and posterior lengths of the testes.

j. Expose the thoracic ganglion (Figure 7), which lies beneath the heart on the floor of the body cavity, by removing the residual hepatopancreas. Remove a 0.5-cm portion of the thoracic ganglion. The correct organ has been obtained if severance of the radiating peripheral nerves causes violent twitching of the respective walking leg of the crab, if the animal has been freshly killed.

k. In female Dungeness crabs, the paired seminal recepticles will be located below and on either side of the thoracic ganglion. Remove the right organ for fixation.

l. Remove both eyestalks and the cerebral ganglion (brain) appearing as a white, pea-sized organ located at the juncture of the eyestalks (Figure 7). This all can be removed as one piece by snipping out with a pair of scissors.

6. All tissues removed from a single crab should be placed into tissue processing cassettes, 4-5 tissue samples to one cassette. Each cassette must be labelled with the animal number from which the tissues were collected. Cassettes are then placed within the sample jar containing fixative.

7. Behavioral, external, and internal abnormalities must be noted on a necropsy field sheet (attached), respectively numbered for a particular pooled crab tissue sample. If no abnormalities within the 20 specimens from a site are observed, then a single field sheet for the sample series will suffice. These field sheets will also contain the label information below and must accompany the samples in a ziploc bag. Be sure to include tissue from a lesion if one is observed--this includes shell lesions as well.

8. A label with crab species, size range and life stage, date of sample, sample location, and contact person's name, address, and telephone number must be placed within each of the sample jars. Use a pencil with soft lead for labelling so that the writing remains legible.

9. Do not mix samples of different crab species within the same jar of fixative. Each species requires a separate jar(s).

10. Place sample jars and ziploc bag containing sample data into a suitable shipping package with adequate packing material to prevent breakage. Plastic jars or containers for fixative and samples work best. Be sure lids are tight and do not leak.

11. Mail to the FRED Division Fish Pathology Lab, [illegible] AK Dept. of Fish and Game, 333 Raspberry Rd., Anchorage, AK 99518-1599; phone (907) 344-0541.

12. Notify the Fish Pathology Lab prior to sample shipment so that samples may be expected and tracked en route.

13. Follow proper procedures and include completed forms regarding chain of custody.

14. Any questions regarding sample preparation should be directed to:

Dr. Ted Meyers
Principal Fish Pathologist III
ADF&G, FRED Division
Juneau Fish Pathology Lab
P.O. Box 3-2000
Juneau, Alaska 99802-2000

Phone: (907) 465-3577

1. Protozoa

Blood Smears

It is essential that slides used in making smear preparations be unscratched, noncorroded, and meticulously clean, free from grease, dust, acid, or alkali; that slides be handled by their edges; that the blood be taken as it exudes, that the process be done rapidly so as to prevent coagulation; and that smears be left to dry in a horizontal position away from flies and dust. The finger tip or structure to be pricked is cleaned with 70% alcohol, after which a prick is made with a blood lancet or a sterilized needle. The first drop is wiped off with absorbent cotton or gauze. Mark necessary data with wax pencil on the end of each slide. Blood films should be stained as soon as possible after drying to insure proper staining.

1. *Thin film*: On slide "A" place a drop of blood about one-half inch from the end. Take a second slide "B" and place it on the surface of the first slide at about a 45° angle, as indicated in Figure 259, and move it to the right until contact is made with the drop of blood. The free end of slide "B" may be supported by the third finger. As soon as it touches the blood, the latter will spread. Now push slide "B" toward the left, being careful to keep the edge pressed uniformly against the surface of slide "A."

In this way a thin smear with uninjured host cells and protozoans and/or microfilariae will be obtained. The size of the drop of blood and acuteness of the angle formed between the slides, will determine the thickness of the film, a more acute angle resulting in a thicker film. Allow film to dry thoroughly.

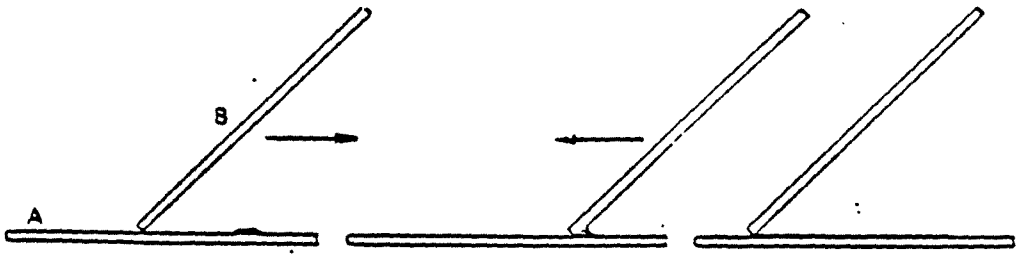

Figure 259. Preparation of thin blood smear.

Fig. 2. The dorsal carapace (cut at B and reflected to the right) is cut below its normal

Fig. 1. The hepatopancreas, testes, and vas deferens have been removed, revealing the anterodorsal heart *1* and the circumesophageal commissures *2*. The large thoracic ganglion *3* lies medioventrally. Its central aperture is visible. In the living animal, the sternal artery passes through the aperture. Connective tissues have been removed in order to show the location of the antennal glands *4*, which lie against the anteroventral face of the exoskeleton.

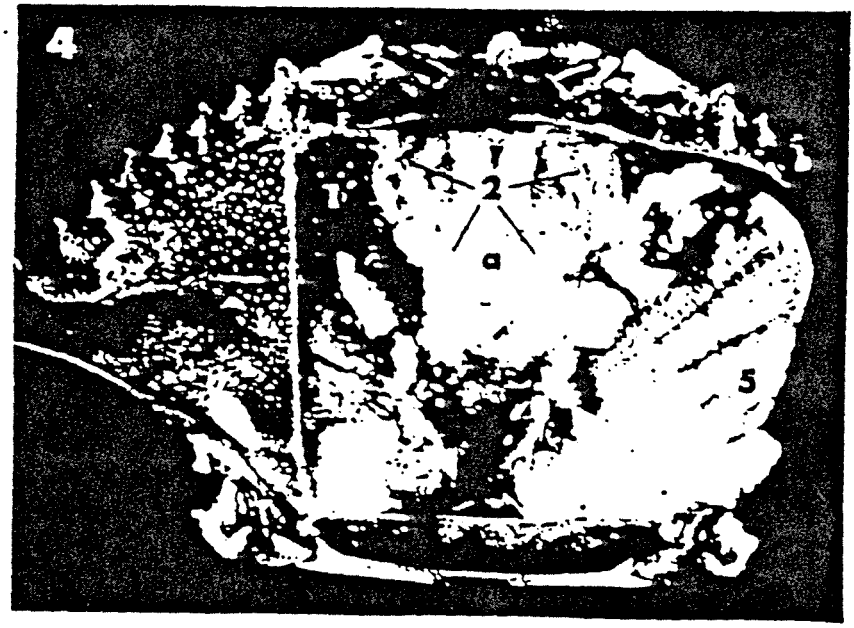

Fig. 4. The medial part of the carapace and much of the epidermis *1* have been removed. The large cardiac stomach *2* is visible. The hepatopancreas tissue lies on the posterior diamond-shaped portion *a*. Two lateral ostia are visible in the semitransparent heart *3*. Intermingled hepatopancreas and testes *4* and the gills *5* can also be seen.

PHOSPHATE BUFFERED FORMALIN

Fixative for Histological Samples of Fish, Bivalves, and Crabs
Histopathology Technical Group

1.	37%-40% Formalin	100.0 ml
2.	Tap water	900.0 ml
3.	$NaH_2PO_4 \cdot H_2O$	4.0 g
4.	Na_2HPO_4	6.0 g

NECROPSY FIELD DATA SHEET FOR HISTOLOGICAL SAMPLES

ADF&G, FRED Division Fish Pathology Lab

Collector/Address/Telephone #

Species

Number Specimens in Sample

Size Range

Life Stage

Date of Collection

Location of Collection (Site Name or Number)

Abnormalities Observed Per Specimen Number

APPENDIX 4: SHRIMP SAMPLING PROCEDURES

ADF&G Prince William Sound Oil Assessment Chain-of-Custody Form

VAL-89-

SERIAL #

Page ____ of ____

Note: Use ballpoint pen, waterproof ink (e.g., Rapidograph) or fine-tip waterproof marker

ADF&G, FRED Division
Fish Pathology Lab
P.O. Box 3-2000
Juneau, AK 99802-2000
Phone: (907) 465-3577

	Date Collected	Sample # (collector's)	Assigned # (leave blank)	Type (tissue, water, sediment, etc.)	Location Collected	Latitude	Longitude	Remarks
5								
10								
15								
20								

(continue on back of page)

CHAIN OF CUSTODY

Samples collected by _____ (print name) of _____ (agency) _____ (signature) _____ (date)

Transferred to _____ (print name) of _____ (agency) at _____ (place) _____ (signature) _____ (date)

Transferred to _____ (print name) of _____ (agency) at _____ (place) _____ (signature) _____ (date)

Transferred to _____ (print name) of _____ (agency) at _____ (place) _____ (signature) _____ (date)

Transferred to _____ (print name) of _____ (agency) at _____ (place) _____ (signature) _____ (date)

Transferred to Ted Meyers of ADF&G at Juneau _____ (signature) _____ (date)

APPENDIX 8: MATERIALS LIST

IRP FIELD SUPPLIES AND EQUIPMENT

Item	Check	Quantity	Vendor and Cat #
Microscope slides			Baxter, #M6146
Syringes - 1 cc			Baxter, #S9501-18
Syringe needles, 22 gx, 1-1/2 in			Baxter, #89549-22J
Syringe needles, 20 gx, 1-1/2 in			
Scissors, 6-1/2 in			Baxter, #D2655-1A
Scissors, 5 in			
Scissors, 4-1/2 in			VWR, #25608-203
Dissecting forceps, 5-1/2 in			VWR, #25718-100
Dissecting forceps, 4-1/2 in			VWR, #25715-043
Specimen forceps, 8 in			
Tissue forceps, 4-1/2 in			Baxter, #D2567-1A
Hemostat, 5 in			Baxter, #D2680-1A
Utility scissors			
Bone cutters			Baxter, #D2576
Scalpel handle, no. 6			
Scalpel handle, no. 5			
Scalpel blades, no. 20			
Scalpel blades, no. 12			
Scalpel blades, no. 10			Baxter, #D2865-10
Dissecting tray			
Tissue cassettes			Baxter, #M7321-33
10% Formalin w/ Na acetate			
Lab markers			
2L Nalgene Bottles			Baxter, #B7541-64
Marker II's			Baxter, #P1220
Slide boxes			VWR, #48450-006
Single-edge blades			
Formalin (37%)			Baxter, #5016-4NY
Sodium Acetate			VWR, #JT3450-11
Prepared 10% buffered formalin			Baxter, #H121-4NY

Project Title: Histopathological Studies of Injury Assessment for Birds, Mammals, Finfish, and Shellfish Exposed to the Oil Spill in Prince William Sound

Brief Flowsheet for Field Sample Collection

1. Collect animals from a specific site.

2. Necropsy.

3. Take tissue samples for the following analyses from the same animal, if possible, or from the same subset in the collection.

   a. Hydrocarbon analysis
   b. Histology
   c. Other (?)

4. Send hydrocarbon sample set to the National Marine Fisheries Service Auke Bay Laboratory in Juneau for storage and later analysis.

5. Send histology sample set to the Alaska Department of Fish and Game, FRED Division repository in Anchorage for storage and later analysis.

6. Other samples, if any, are sent to their respective storage site for later analysis.

7. Don't forget chain-of-custody paperwork to accompany each sample set.

Qualifications of Project Leader

CHARLES E. O'CLAIR S.S. No.: 012-32-1851

Personal: Born May 29, 1941; Ayer, Massachusetts

Education: University of Massachusetts, B.S., Zoology, 1963
University of Washington, Ph.D., Fisheries, 1977

Experience: 1977 - present: Fishery Biologist (Research), National Marine Fisheries Service, Auke Bay Laboratory, Juneau, Alaska. Research experience includes five years of field and laboratory work on the effects of oil pollution on benthic invertebrates in conjunction with the Outer Continental Shelf Energy Assessment Program followed by seven years of research on the ecology and behavior of Dungeness, king, and tanner crab in relation to the management of these species.

1983-1987: Affiliate Assistant Professor of Fisheries, School of Fisheries and Science, University of Alaska, Juneau.

Selected Publications:

O'Clair, C. E. and S. D. Rice. 1985. Depression of feeding and growth rates of the seastar Evasterias troschelii during long-term exposure to the water-soluble fraction of crude oil. Mar. Biol. 84:331-340.

O'Clair, C. E. and S. T. Zimmerman. 1987. Biogeography and ecology of intertidal and shallow subtidal communities. pp. 305-344. In The Gulf of Alaska: Physical Environment and Biological Resources. D. W. Hood and S. T. Zimmerman, Eds. National Technical Information Service, Springfield, Virginia.

O'Clair, C. E. and J. L. Freese. 1988. Reproductive condition of Dungeness crabs, Cancer magister, at or near Log Transfer Facilities in Southeastern Alaska. Marine Environ. Res. 26:57-81.

Morado, J. F., A. K. Sparks and C. E. O'Clair. 1988. A preliminary study of idiopathic lesions in the Dungeness crab, Cancer magister, from Rowan Bay, Alaska. Marine Environ. Res. 26:311-318.

O'Clair, C. E., R. P. Stone and J. L. Freese. (in press). Movements and habitat use of Dungeness crab and the Glacier Bay fishery. Proc. Second Glacier Bay Science Symposium, 1988.

CONFIDENTIAL

DRAFT

STATE/FEDERAL NATURAL RESOURCE DAMAGE ASSESSMENT
DETAILED STUDY PLAN

Project Title:	INJURY TO PRINCE WILLIAM SOUND SPOT SHRIMP
Study ID Number:	Fish/Shellfish Study Number 15
Lead Agency:	State of Alaska, ADF&G, Commercial Fisheries Division
Cooperating Agency(ies):	None
Principle Investigator:	Charles Trowbridge, Fisheries Biologist
Assisting Personnel:	Wayne Donaldson, Fisheries Biologist John Hilsinger, Fisheries Biologist Fisheries Technician (1)
Date Submitted:	October 12, 1989

	Signature	Date
Principle Investigator:	Charles E Trowbridge	10-10-89
Supervisor:	Wayne Donaldson	10-10-89
OSIAR Senior Biometrician:	W/dr	
OSIAR Program Manager:	[illegible]	10-13-89
OSIAR Director:	Michael Rieser	10-17-89

INTRODUCTION

This project is aimed at assessing possible damage to spot shrimp, Pandalus platyceros, due to Prudhoe Bay crude oil spilled from the Exxon Valdez. Spot shrimp are a commercially important species and also support subsistence and personal use fisheries in Prince William Sound.

Spot shrimp are known to be sensitive to oil contamination. To determine the impacts that hydrocarbons from the spill may have had on spot shrimp, samples will be collected from three oiled and three non-oiled sites in western Prince William Sound. The data collected from the samples will be analyzed to determine tissue hydrocarbon levels and tissue damage. The collected data will also be tested to confirm or reject the hypothesis that there is no significant difference between the oiled and non-oiled areas. Relative abundance at each study site and changes in relative abundance over time will be tested to determine if those changes are correlated to the presence of oil. The size composition of the stock at each site will be determined and analyzed to see if the 1989 year class suffered a high mortality rate in areas of high oil impact relative to other year classes in oiled and non-oiled areas. Spot shrimp fecundity will also be determined and tested to see if there are significant differences between oiled and non-oiled sites.

OBJECTIVES

1. Measure hydrocarbon concentrations in spot shrimp from oiled and non-oiled areas of Prince William Sound and test the hypothesis that the level of hydrocarbons is not related to the level of oil contamination present at a site. The experiment is designed to detect a difference of 1.2 standard deviations in hydrocarbon content with the probability of making a type I or type II error of 0.05 and 0.10, respectively.

2. Determine the relative abundance of spot shrimp in oiled and non-oiled areas.

3. Analyze egg fecundity, mortality, and sublethal effects of oil for oiled and non-oiled areas, and determine whether those effects result in adverse changes in viability.

4. Identify potential alternative methods and strategies for restoration of lost use, populations, or habitat where injury is identified.

METHODS/DATA ANALYSIS

This project uses standardized commercial spot shrimp pots (Appendix G) to catch spot shrimp in oiled and non-oiled areas. Fishing will be carried out by the ADF&G research vessel Montague. Shrimp specimens will be analyzed for Prudhoe Bay crude oil levels and necropsied to determine if damage has occurred to tissues due to oil contamination. Oiled and non-oiled areas will be sampled in two phases which correspond with two stages of egg development. The first phase will occur in early November following the fall molt and egg extrusion. The second phase will occur in early March just prior to egg hatching. The sampling strategy will be identical during both phases. Relative abundance estimates of spot shrimp will be made using a stratified pot deployment based on depth and location. Size distribution, species composition, and reproductive data will also be collected. Previous spot shrimp research in Prince William Sound is documented by Kimker and Donaldson (1987), Donaldson (1989), Donaldson and Trowbridge (1989), and Kruse and Murphy (1989).

Study Sites

This project will be carried out in two general areas. One will be an area of little apparent impact, the northwestern portion of Prince William Sound. This area includes Unakwik Inlet, the site of previous ADF&G research on abundance and growth of spot shrimp. The second area will be central and southwestern Prince William Sound, an area of generally high oil impact. This area includes Green Island where ADF&G test fishing occurred in 1981.

Within each of these two areas, test fishing will take place at three sites. In the northwestern sound, test fishing will occur in Unakwik Inlet, Port Wells, and Culross Passage. In the central and southwestern sound, test fishing will take place near Eleanor Island, Knight Island Passage, and Green Island (Appendix B; Figure 1). Shrimp distribution in these areas will be established by surveying the commercial fleet.

Sample Design

Fishing will take place at 6 sites - 3 in oiled areas and 3 in non-oiled areas (Appendix B, I.). Each site will be stratified by depth. Stratum 1 will be shallow waters - 20 to 70. Stratum 2 will be deep waters - 70 to 120. Based on past research, spot shrimp are not abundant below those depth ranges. Because of the difficulty of placing the gear at precise depths, it is impractical to divide the depth into more than two strata. Strata span 50 fathoms in depth or approximately 65 to 85 fathoms in width along the bottom at slopes of 75 to 100 percent (Appendix B, Figure 2). Fishing 11 pots on a 50 fathom string will span the width of each strata and allow for a complete placement of gear over the strata.

Eleven pots spaced 5 fathoms apart will be fished on a long line so that each string of pots is 50 fathoms long. One 50 fathom string of gear constitutes a sampling station. Two stations will be fished in each stratum at each site for a total of 22 pots per stratum per site, or 44 pots per site. Forty-four pots is the most that can be fished in a day while collecting all of the various samples and data (Appendix B, IV.). A different site will be sampled each day. A total of 264 pots will be fished during each time period.

Data Collection

Station information including location (latitude and longitude), depth in fathoms, time and duration during which pots fish will be recorded by the vessel skipper on a paper form (Appendix A, Figure 1).

Environmental Samples:

Water temperature, salinity, and dissolved oxygen concentration by depth will be recorded using a Sea Bird Electronics CTD, transferred from the CTD to a micro-computer and stored on diskette. CTD casts will be at one station in the deep stratum every day. The CTD will be let down at a rate of 60 meters per minute. Because of the configuration of the CTD, only readings from the downcast will be used.

Biological Samples:

Total weight of catch, sub-sample weight, and the weight of each species in a sub-sample will be recorded for each pot on a paper form at the time the pot is retrieved (Appendix A, Figure 2). The total weight of shrimp per pot will be determined by weighing the contents of each pot on a spring scale. If the pot contents are less than 1 kilogram of shrimp the entire shrimp catch will be processed. If the pot content exceeds 1 kilogram of shrimp, a 1 kilogram sub-sample of shrimp will be obtained by scooping out 1 kilogram of shrimp at random.

Each sub-sample will be sorted by species. Weight and number of animals will be recorded for each species. Only spot shrimp will be retained for further data collection. All spot shrimp in the sub-sample will be measured for carapace length in millimeters using a digital caliper and sex will be determined as male, transitional, or female according to the methods described in Appendix C. For female spot shrimp, egg color and stage of development (eyed or uneyed); relative clutch size; presence of breeding dress and egg parasites

or parasitic externa will be noted. The data collected will be recorded on a paper data form (Appendix A, Figure 2).

For female spot shrimp only, fecundity will be determined from samples returned to the Cordova laboratory, recorded on a paper form, and entered into an R:base data base.

Necropsy Samples:

Specimens for necropsy analysis will be taken after the catch is weighed and processed. Twenty shrimp from a single station in each stratum will be selected randomly to make up a necropsy sample. These samples will be taken according to the methods specified by ADF&G FRED Division pathologist Ted Meyers (Appendix D). Necropsy samples will be labeled with the date, station number, latitude and longitude, sample number, project leader's name, species, and agency (Appendix A, Figure 3).

Hydrocarbon Samples:

To prevent contamination specimens for hydrocarbon testing will be taken from the pot immediately after removal from water and before contents are weighed. Three spot shrimp will form one composite sample. Each composite will be taken from a different pot. Two replicates of the composite will be taken randomly from one station in the stratum and the third replicate will come from the other station. Three samples per site per depth stratum result in 9 samples per depth stratum (3 sites X 3 samples) per impact level and 18 samples per oil impact level (9 samples X 2 depth strata). This will allow hypothesis testing to detect differences in hydrocarbon levels of 1.2 standard deviations with the probability of a type I or type II error being 0.05 and 0.10, respectively.

The number of specimens for one hydrocarbon analysis is dependent on the size of the specimens collected. Tissue volume based on the average size of the species was estimated and the number of specimens needed to provide 15 gm of tissue was calculated to be three spot shrimp. An estimate of 3 hydrocarbon samples from each treatment level is needed for detecting contamination between levels (Dr. B. Clark, personal communication).

All hydrocarbon samples will processed according to the methods specified by NOAA (Appendix E). Samples will be labeled with the date, station number, latitude and longitude, sample number, project leader's name, species, and agency (Appendix A, Figure 3). Hydrocarbon and necropsy samples will be sealed with evidence tape, kept in secured locations, and chain of custody will be maintained according to the methods described in Appendix F.

Fecundity samples:

Twenty five egg bearing females will be taken at random from each station to estimate fecundity and egg mortality. A total of 24 stations will yield a total sample size of 600 females. Specimens from each station will be placed together in a plastic bag in whole condition. Each sample bag will be labeled with project leader's name, species name, "eggs", date, station, and agency name.

Fecundity will be determined by removing the eggs from the pleopods, drying each egg mass to a constant weight, weighing a sub-sample of a known number of eggs, and expanding the sub-sample weight to the weight of the entire clutch. Carapace length will be taken for each specimen at the time the eggs are removed and recorded on the fecundity form (Appendix H).

The number of shrimp sub-sampled for fecundity estimation will be determined by time and budget constraints. If not all 25 shrimp from the station samples can be processed due to the above constraints, sub-samples of a smaller but equal number will be processed for each station. If possible additional numbers from the remainder will be processed at a later time. A minimum number of five shrimp from each station will be sampled for fecundity which will allow an adequate sample (thirty per depth strata per oil impact level) to test for differences in fecundity between depth strata and oil impact level.

Data analysis

To address objective 1, the average levels of Prudhoe Bay crude oil present in spot shrimp tissue by strata and site will be determined. Significant differences in hydrocarbon concentrations between oiled and non-oiled sites will be tested for using analysis of variance. To further define the impact of hydrocarbon levels on the stock, the percentage of animals with abnormal tissues in oiled and non-oiled areas will be determined. A chi-square test will be utilized to test for significant differences in percentage of animals with abnormal tissues between strata, sites, and impact levels.

Objective 2 will be addressed by determining the average catch per pot by weight, sex, and species. Analysis of variance will be used to test for significant differences in each of these categories between strata (depth), sites, and oiled versus non-oiled areas. To define the relationship between hydrocarbon levels and changes in relative abundance, statistics for analysis of covariance or an appropriate multivariate technique will be calculated to contrast differences in hydrocarbon content and relative abundance in oiled and non-oiled areas. Changes in average catch per pot over time will also be analyzed between different depth strata, sites, and oiled and non-oiled areas.

A size frequency distribution will be made by species and sex. The

hypothesis that there is no significant difference between strata, and oil impact levels for size frequency distribution will by tested using a t test or a similar non-parametric test. Changes in size frequency distribution over time will be examined by comparing data collected during phase one and phase two. A t test will be used to look for significant differences between time periods as well.

To meet objective 3, the relationship between size and fecundity will be examined. The percentage of spot shrimp females bearing eggs; the stage of spot shrimp egg development (color and presence or absence of eyes); the percentage of spot shrimp egg fouling and egg mortality; the fecundity by size; and the relative clutch size will be determined for each station and each phase. Chi-square tests will be used to test for differences in strata, sites and levels in data which involve percentages and proportions. Differences between strata, sites, and impact levels for fecundity and relative size of clutch will be tested for using analysis of variance. ANOVA will also be used to test for a significant difference in the above measures between phase one and phase two which may provide an estimate of the number of eggs dying over the course of the brood period or estimates of differences in egg viability.

To meet objective 4, and to identify potential methods or strategies for restoring this population if it is damaged by oil, it will be necessary to test changes in catch per unit effort, age class strength, and reproductive viability to see if management actions implemented to restore damaged stocks are having the desired effect. All data will be analyzed for evidence of damage. Depending on the degree of damage to the stock, fishery management actions may be necessary to reduce additional mortality to an oil damaged stock. This may include fishery restrictions, total closure or other measures. Additionally, the need for continual study and monitoring of stock condition will be assessed.

All catch, size, and station data will be entered into R:base applications onboard ship using portable micro computers. Final analyses will take place in the Cordova and Anchorage offices using R:base on micro computers. Data mapping will be done using Sea Plot software (which is a specialized R:base application). Fecundity versus size relationships will be determined using Stats Plus software. Statistical tests will be conducted in Juneau, Cordova, or Anchorage using micro-computers running Stats Plus, SAS, Minitab, or Systat software.

SCHEDULES AND REPORTS

Date(s)	Activity
November 1989	Phase 1 field program will last approximately 7 days. (Approximately Nov. 1 - 7, 1989); Sampling will occur daily while in the field. One of the six strata will be sampled each day, day one will be used for travel to the area and setting the initial 2 strings of pots.
November 1989	Laboratory analysis, hydrocarbon analysis.
November-December 1989	Data entry and analysis.
December 1989	Preliminary report on impacts of oil on shrimp.

PROJECT BUDGET

Line Item	Category	Budget
100	Personal Services	$ 25,400
200	Travel	$ 1,500
300	Contractual	$ 20,300
400	Commodities	$ 2,300
500	Equipment	$ 11,000
Total		$ 60,500

FUNDED PERSONNEL

Class	PCN	PFT_mm	SFT_mm
FB III	11-1084	2	
FB I	11-1649		3
FT II	11- new		2

LITERATURE CITED

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APPENDIX A, FIGURE 1

ADF&G SPOT SHRIMP SURVEY
PWS OIL SPILL IMPACT ASSESSMENT PROJECT
STATION DATA FORM

VESSEL________________________________ CRUISE ☐☐☐ DATE ☐☐☐☐☐☐

SITE______________ STRATUM ☐ LAT. ☐☐ ☐☐☐ LONG. ☐☐☐ ☐☐☐

STRING ☐ DEPTH RANGE ☐☐☐ - ☐☐☐ NUMBER OF POTS ☐☐

TIME SET ☐☐☐☐ TIME PULLED ☐☐☐☐ DATE PULLED ☐☐☐☐ SOAK TIME ☐☐

WEATHER__________________________ SEA_______________ SWELL____________

NOTES____

____

SITE______________ STRATUM ☐ LAT. ☐☐ ☐☐☐ LONG. ☐☐☐ ☐☐☐

STRING ☐ DEPTH RANGE ☐☐☐ - ☐☐☐ NUMBER OF POTS ☐☐

TIME SET ☐☐☐☐ TIME PULLED ☐☐☐☐ DATE PULLED ☐☐☐☐ SOAK TIME ☐☐

WEATHER__________________________ SEA_______________ SWELL____________

NOTES____

____

SITE______________ STRATUM ☐ LAT. ☐☐ ☐☐☐ LONG. ☐☐☐ ☐☐☐

STRING ☐ DEPTH RANGE ☐☐☐ - ☐☐☐ NUMBER OF POTS ☐☐

TIME SET ☐☐☐☐ TIME PULLED ☐☐☐☐ DATE PULLED ☐☐☐☐ SOAK TIME ☐☐

WEATHER__________________________ SEA_______________ SWELL____________

NOTES____

____

SITE______________ STRATUM ☐ LAT. ☐☐ ☐☐☐ LONG. ☐☐☐ ☐☐☐

STRING ☐ DEPTH RANGE ☐☐☐ - ☐☐☐ NUMBER OF POTS ☐☐

TIME SET ☐☐☐☐ TIME PULLED ☐☐☐☐ DATE PULLED ☐☐☐☐ SOAK TIME ☐☐

WEATHER__________________________ SEA_______________ SWELL____________

NOTES____

____

APPENDIX A, FIGURE 2

ADF&G SPOT SHRIMP POT SURVEY
PWS OIL IMPACT ASSESSMENT PROJECT
SHRIMP DATA FORM

VESSEL ____________________ CRUISE ☐☐☐ DATE ☐☐☐☐☐☐

SITE ____________ STRATUM ☐ STRING ☐ POT NO. ☐☐ NOTES ________

TOTAL SHRIMP WT. ☐☐☐☐☐ SUBSAMPLE WT. ☐☐☐☐ SAMPLE FRACTION ☐☐

SPOTS (5) WT. ☐☐☐ NO. ☐☐☐

PINKS (2) WT. ☐☐☐ NO. ☐☐☐

COONSTRIPES (3) WT. ☐☐☐ NO. ☐☐☐

SPE-CIES	SEX	CAR. LEN.	B.D.	EGGS			PAR	SPE-CIES	SEX	CAR. LEN.	B.D.	EGGS			PAR
				CON	COL	FOU						CON	COL	FOU	

APPENDIX A, FIGURE 3

ADF&G SPOT SHRIMP POT SURVEY
PWS OIL IMPACT ASSESSMENT PROJECT
HYDROCARBON AND NECROPSY SAMPLE FORM

VESSEL_____ CRUISE ☐☐☐ DATE ☐☐☐☐☐☐

- SAMPLE COLLECTION -
CHECK BOX IF SAMPLE WAS COLLECTED AND INDICATE SAMPLE #

SITE_____ STRATUM ☐ LAT. ☐☐ ☐☐☐ LONG. ☐☐☐ ☐☐☐

DEPTH RANGE ☐☐☐-☐☐☐ NOTES_____

REP. 1 HYDROCARBON ☐ SAMPLE NO._____

REP. 2 HYDROCARBON ☐ SAMPLE NO._____

REP. 3 HYDROCARBON ☐ SAMPLE NO._____

NECROPSY ☐ SAMPLE NO._____

SITE_____ STRATUM ☐ LAT. ☐☐ ☐☐☐ LONG. ☐☐☐ ☐☐☐

DEPTH RANGE ☐☐☐-☐☐☐ NOTES_____

REP. 1 HYDROCARBON ☐ SAMPLE NO._____

REP. 2 HYDROCARBON ☐ SAMPLE NO._____

REP. 3 HYDROCARBON ☐ SAMPLE NO._____

NECROPSY ☐ SAMPLE NO._____

APPENDIX A. Table 1

CODES FOR SPOT SHRIMP FORMS

Stratum

1 = Shallow (20 - 70 fathoms)
2 = Deep (70 - 120 fathoms)

Species

1 = Sidestripes (Pandalopsis dispar)
2 = Pinks (Pandalus borealis)
3 = Coonstripes (Pandalus hysinotus)
4 = Humpies (Pandalus goniuris)
5 = Spots (Pandalus platyceros)

Sex

1 = Male
2 = Female
3 = Transitional
4 = Sex Not Determined

Breeding Dress (Females Without Eggs Only)

1 = No
2 = Yes

Egg Condition

0 = No eggs
1 = Eyed Eggs
2 = Uneyed Eggs

Egg Color

0 = Light Orange
1 = Dark Orange
2 = Yellow
3 = Light Brown
4 = Dark Brown
5 = Blue

Egg Fouling

1 = Absent
2 = Present

Parasites

1 = Egg Predator
2 = Parasitic Externa
3 = Carapace Parasite

APPENDIX B.

ADF&G SPOT SHRIMP SAMPLING PLAN

I. SITES

A. Non-oiled

1. Unakwik Inlet
2. Port Wells
3. Culross Pass

B. Oiled

1. Eleanor Island
2. Knight Island
3. Green Island

C. Areas are shown on Appendix B. Figure 1.

II. STRATA

A. Shallow stratum will be from 20 to 70 fathoms.

B. Deep stratum will be from 70 to 120 fathoms.

C. Strata depth ranges were chosen based on known distribution of spot shrimp, the desire to sample all depth ranges where spot shrimp are known to be abundant, and the fact that 11 pots per longline is a manageable number of pots to fish per line, while still having coverage of all depths.

III. STATIONS

A. Exact station locations at each site are being chosen with the help of fishermen experienced at spot shrimp fishing in those areas. Stations will not be selected until immediately before the sampling begins in order to allow the greatest amount of input possible.

B. Each station will consist of one string of eleven pots fished on a long line. Pots will be spaced 5 fathoms (30 feet apart) for a total length of 50 fathoms for each string of pots.

C. There will be two stations in each stratum. These two stations will be fished on the same day.

IV. FISHING PLAN

A. Weekly Schedule

1. Day 1 — Sail to Unakwik Inlet set stations 1 - 4.

   Day 2 — Pick Stations 1 through 4. Sail to Port Wells and set stations 5 - 8.

   Day 3 — Pick stations 5 through 8. Sail to Culross Passage and set stations 9 - 12.

   Day 4 — Pick stations 9 through 12. Sail to Eleanor Island and set stations 13 - 16.

   Day 5 — Pick stations 13 through 16. Sail to Knight Island and set stations 17 - 20.

   Day 6 — Pick stations 17 through 20. Sail to Green Island and set stations 21 - 24.

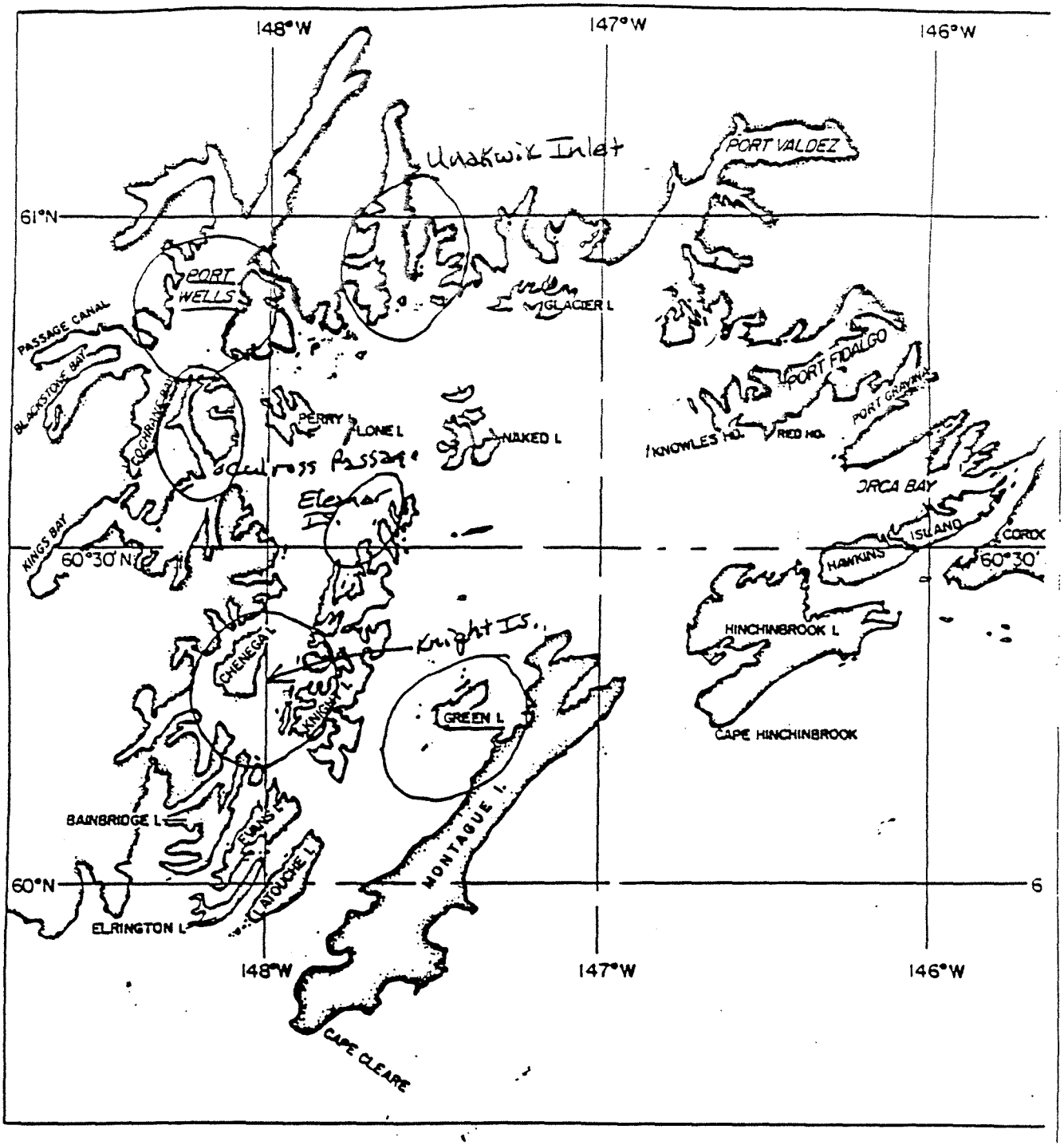
   Day 7 — Pick stations 21 through 24. Sail to Cordova. End of trip.

B. Daily Schedule

1. Gear will fish a standardized overnight period of 20 to 24 hours.

2. Pots will be pulled in the morning and subsequently set such that the desired soak time will be achieved. If the desired soak time cannot be achieved, pots will be fished to minimize variance from this desired fishing time.

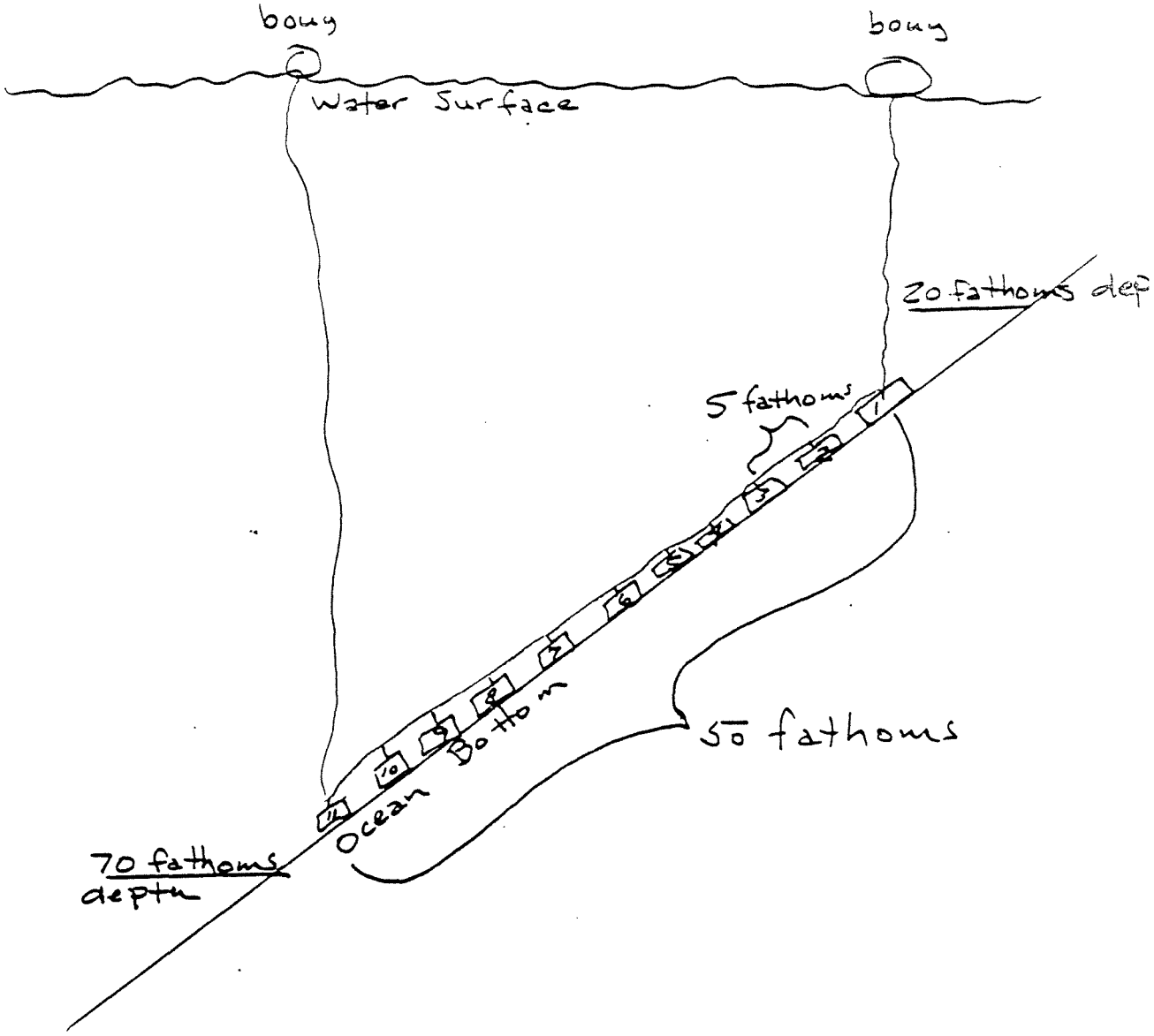
Appendix B. Figure 1



spot shrimp survey sites in Prince William Sound

APPENDIX B. Figure 2

SPOT SHRIMP POT DEPLOYMENT



APPENDIX C

STANDARD OPERATING PROCEDURE FOR SEX DETERMINATION OF PANDALID SHRIMPS IN PRINCE WILLIAM SOUND

by

Charlie Trowbridge

October 10, 1989

Determining the sex of a Pandalid shrimp is best accomplished using secondary sexual characteristics. Sexing based upon primary sex organs is difficult and time consuming. Using a secondary sex characteristic such as endopod development, which closely tracks the gonad development, allows sex to be determined with relative ease. The sex of large Pandalids such as P. platyceros can be found by visual inspection, without aid of magnification.

Materials used in sexing are a sharp needle probe, a good light preferably with a dark background, and a source of magnification, two to seven power (2x-7x) in strength in order to clearly view endopod characteristics. A visor with two-power optics in place has worked well however a stronger magnification would be helpful for small specimens.

Pandalid shrimp in Alaska are typically protandric hermaphrodites. Therefore three sexual stages can be identified, male, female, and transitional. Sexing of Pandalid shrimp in Prince William Sound is performed according to Butler's description in his work Shrimps of the Pacific Coast of Canada (1980).

The size of a shrimp may give some indication of its sex eg. a small specimen would probably be a male and a large specimen a female. The endopod of the first pleopod is first inspected. If the endopod terminates in two rounded lobes approximately equal in length it is a male. Depending upon specimen size and magnification a small clump of "hooklike setae" may be visible on the inner lobe. A female will be indicated by the endopod of the first pleopod terminating in a single firmly-pointed lobe. A transitional could be described as being intermediate between the characteristics of the male and the female in that the two lobes are still present however the inner lobe is shrunken to form a small stiff appendage which may be hidden along the inner margin of the endopod. The outer lobe of the endopod in the transitional is usually somewhat larger, firmer, and more pointed than the male stage.

The second pleopod may also be inspected to determine sex and Butler cautions that for a neophyte this is the characteristic to use first. A male can be identified as having two small processes

nearly the same length branching from the medial end of the endopod. The inner process is the appendix masculina and will have spines along its tip. The outer process is the appendix interna which will be tipped with "hooklike setae". In the female only the appendix interna will be present. A transitional may be identified as having both processes with the appendix masculina being approximately one-half (or less) the length of the appendix interna.

Allen (1959) gives a detailed account of these same morphological changes which aid in determination of sex in Pandalus borealis. Drawings in his paper are more extensive than those in Butler but both authors appear to agree on the use of endopods in sexing Pandalid shrimp.

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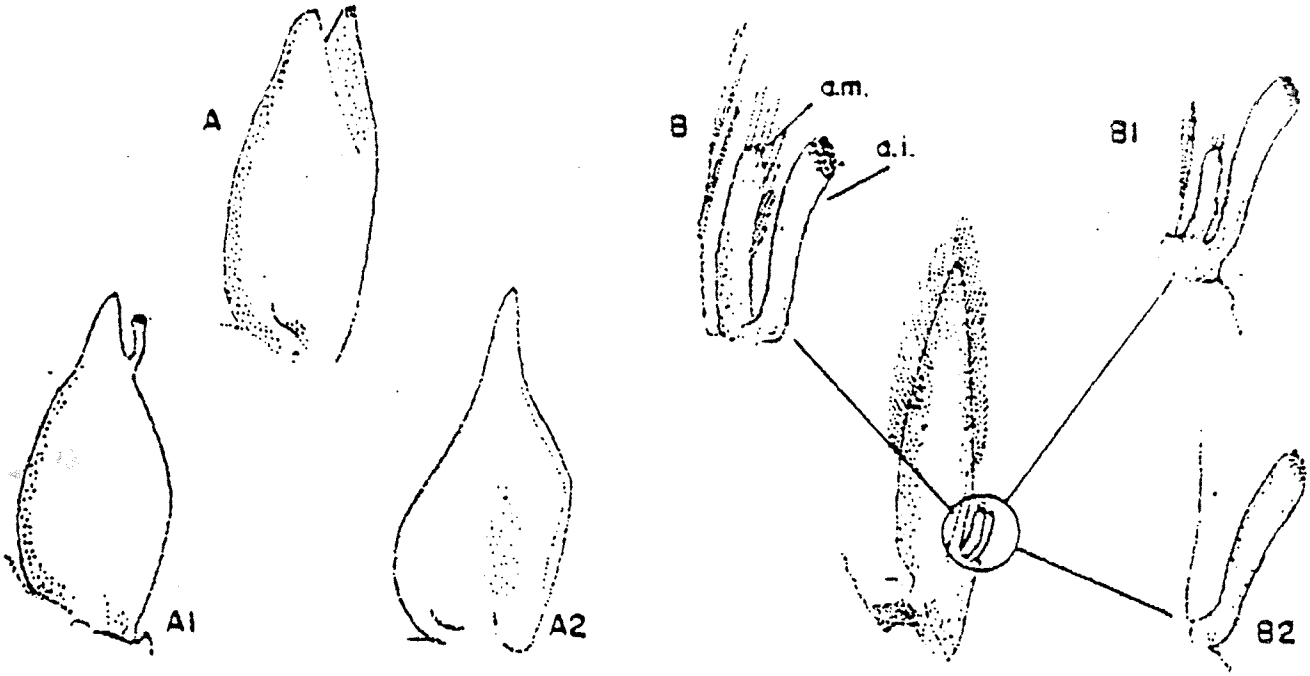


FIG. 1. *Pandalopsis dispar*. Endopod of first pleopod: (A) active male phase. (A_1) transitional phase. (A_2) female phase. Endopod of second pleopod: (B) active male phase. (B_1) transitional phase. (B_2) female phase. a.i., appendix interna; a.m., appendix masculina. (from Butler, 1980)

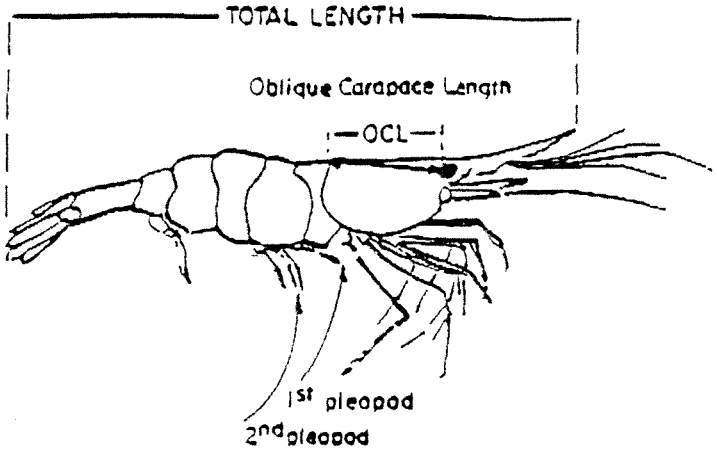


Figure 9.—Method of measuring the carapace of *Pandalus borealis* and location of the first and second pleopods.

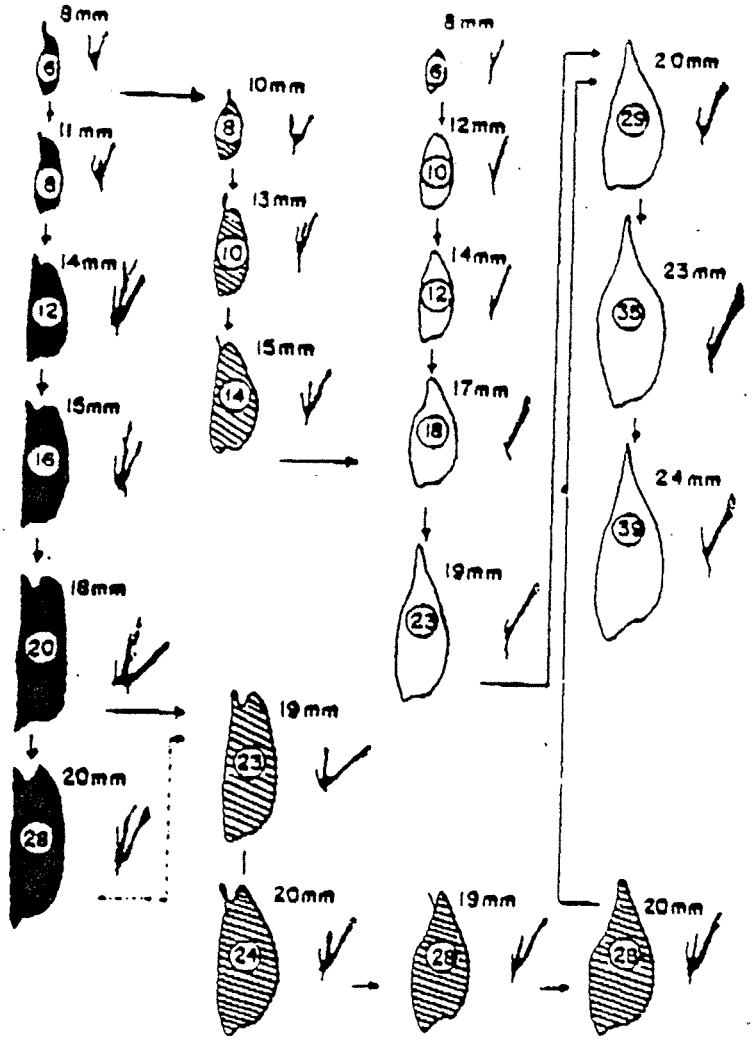


Figure 10.—Changes in form with increasing age of the endopodite of the first pleopod and the corresponding appendix interna and appendix masculina of the second pleopod of *Pandalus borealis* from the Northumberland population. Age in months is given in the ring in each endopodite and the carapace length (mm) above each figure. Male endopodite, black; transitional, cross-hatched; female, outlined. Arrows indicate sequence (from Allen 1959).

APPENDIX D

HISTOLOGICAL SAMPLE PREPARATION FOR SHRIMP

Histopathology Technical Group
(Taken from Bell and Lightner 1988)

NOTE: Only live or moribund shrimp will be suitable for processing. Histopathological changes caused by toxic chemicals are often very subtle at best. Tissues in dead shrimp autolyze very quickly and will mask these changes. Do not collect and process dead shrimp. Keep shrimp alive in containers of seawater if they must be transported to the processing site. Also, minimize the handling stress on the live shrimp to be preserved so that stress-mediated histological artifacts do not occur. Do not over-ice animals such that tissues freeze while in transit. Frozen tissues are worthless for histological examination.

1. The fixative to be used is 10% neutral buffered formalin (formula attached). Formalin should be handled wearing rubber or latex gloves.

2. The volume of fixative should be ten times the volume of the tissue. This is important since any less fixative may result in tissue autolysis and worthless samples. After 72 hours, shrimp specimens should be transferred to 70% ethyl alcohol for shipment and storage. This prevents tissues from becoming too hard and brittle when stored in fixative for long periods. Also, the fixative poured off may be saved and strained of tissue fragments and used one more time for other samples.

3. The sample size per site or species will be 20 shrimp, live or moribund.

4. The chitinous exoskeleton of shrimp prevents adequate penetration of any fixative by simple immersion. Consequently, the fixative must be injected into strategic internal areas of each animal prior to dropping the whole shrimp into the fixative. Inject fixative into the living shrimp using a 10-ml syringe and appropriately sized needle, depending upon the size of the animal (small shrimp; i.e., small-gauge needle). This procedure is described by the following:

   a. First inject laterally into the hepatopancreas; i.e., cephalothorax region (Figure 1a).
   b. Then inject dorsally into the region anterior to the hepatopancreas; i.e., between the thorax and the eyestalks (Figure 1b).
   c. Inject the posterior abdominal region (Figure 1c).
   d. Inject the anterior abdominal region (Figure 1d).

   Inject more of the fixative into the hepatopancreas than the other sites but overall use about 5%-10% of the shrimp's body weight. All signs of life should disappear.

   e. Immediately after injection, slit the cuticle of the animal from the last (6th) abdominal segment to the base of the rostrum. The incision in the cephalothoracic region should be just lateral to the dorsal midline and that in the abdominal region should be mid-lateral (Figure 2). Do not cut too

deeply into the underlying tissue. The objective is to break the cuticle to allow fixative penetration.

SHRIMP LARGER THAN 12 GRAMS

a. Same as above, but a transverse cut should be made at the abdomen/cephalothorax junction (Figure 3a) and again midway across the abdominal area (Figure 3b).

5. After injection and body incisions, the animal may be dropped into the fixative.

6. External abnormalities and unusual behavior must be noted on a necropsy field data sheet (attached), respectively numbered for a particular specimen in the sample. If no abnormalities are observed within the 20 specimens from a site, then a single field sheet will suffice for the sample series. These field sheets will also contain the label information below and must accompany the samples in a ziploc bag.

7. A label with shrimp species, size range and life stage, date of sample, location of sample, and contact person's name, address, and telephone number must be placed within each of the sample jars. Use a pencil with soft lead for labelling so that the writing remains legible.

8. Do not mix samples of different shrimp species within the same jar of fixative. Each species requires a separate jar(s).

9. Place sample jars and ziploc bag containing the sample data into a suitable shipping package with adequate packing material to prevent breakage. Plastic jars or containers for fixative and samples work best. Be sure lids are tight and do not leak.

10. Mail to the FRED Division Fish Pathology Lab, P.O. Box 3-2000, Juneau, Alaska, 99802-2000; phone: (907) 465-3577.

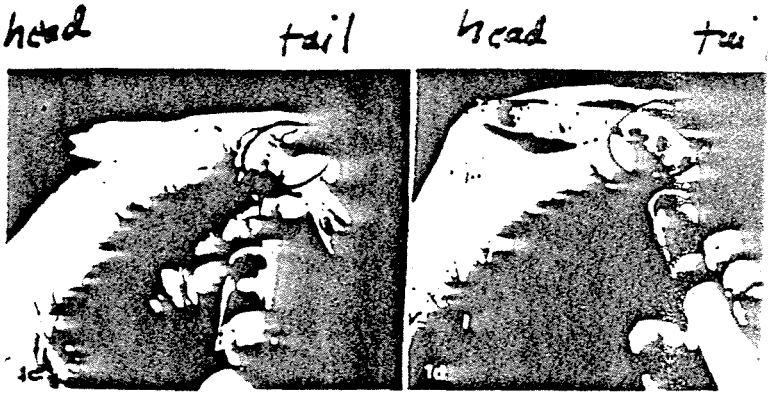
11. Notify the Fish Pathology Lab prior to sample shipment so that samples may be expected and tracked en route.

12. Follow proper procedures and include completed forms regarding chain of custody.

13. Any questions regarding sample preparation should be directed to:

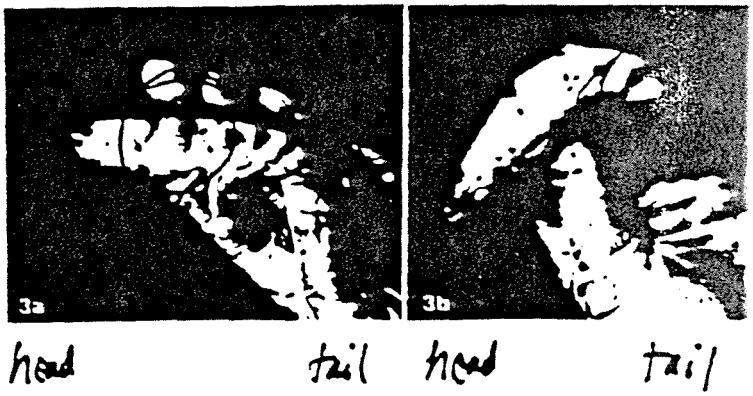
Dr. Ted Meyers
Principal Fish Pathologist III
ADF&G, FRED Division
Juneau Fish Pathology Lab
P.O. Box 3-2000
Juneau, Alaska 99802-2000

Phone: (907) 465-3577



4) Immediately following injection, slit the cuticle, with disse scissors, from the sixth abdominal segment to the base o rostrum, paying particular attention not to cut deeply int underlying tissue. The incision in the cephalothoracic r should be just lateral to the dorsal midline, while that i abdominal region should be approximately mid-lateral (Figu

5) Shrimp larger than 12 grams, should then be transversely slit at the abdomen/cephalothorax junction (Figure 3a) or again abdominally (Figure 3b).
6) Following injection, incisions and bisection/trisection, imm the specimen in the remainder of the fixative.



PHOSPHATE BUFFERED FORMALIN

Fixative for Histological Samples of Fish, Bivalves, and Crabs
Histopathology Technical Group

1.	37%-40% Formalin	100.0 ml
2.	Tap water	900.0 ml
3.	$NaH_2PO_4 \cdot H_2O$	4.0 g
4.	Na_2HPO_4	6.0 g

NECROPSY FIELD DATA SHEET FOR HISTOLOGICAL SAMPLES
ADF&G, FRED Division Fish Pathology Lab

Collector/Address/Telephone #

Species

Number Specimens in Sample

Size Range

Life Stage

Date of Collection

Location of Collection (Site Name or Number)

Abnormalities Observed Per Specimen Number

Appendix E

PROCEDURES FOR TAKING HYDROCARBON AND HISTOLOGY SAMPLES MULTI-SPECIES TRAWL SURVEY, PRINCE WILLIAM SOUND

The following procedures should be used in taking hydrocarbon and histology samples during trawl surveys in Prince William Sound. The hydrocarbon and histology procedures were developed during the May/June 1989 trawl survey. The procedures developed at that time have been modified slightly based on subsequent information on taking samples and proper custody of the samples. Two handouts should be consulted prior to taking the samples, 1) Histopathology Technical Group for Oil Spill Assessment Studies in Prince William Sound, Alaska, and 2) Chain-of-Custody Procedures. The handouts are appended to these Procedures.

Equipment

1. Stainless steel pans, with lids: three assorted sizes for holding and storing knives, scalpels, and scissors.
2. Paper towels.
3. Scalpels: four styles of Bard Parker blades: #10 deep belly, larger deep belly, #11 straight point, and #12 hooked. For cutting tissue and puncturing bile gland.
4. Short blade boning knives: two. For incising and cutting tissue.
5. Long, thin blade fish fillet knives: two. For incising and cutting tissue.
6. Scissors: surgical type with thin, pointed blade and larger, heavy duty type. For cutting tissue and exoskeleton.
7. Forceps: three sizes, small to large. For holding delicate tissue up to large visceral mass.
8. Methylene chloride (MC) for rinsing equipment.
9. Teflon squeeze bottles for using the MC.
10. Steel for knife sharpening.
11. Surgical gloves.
12. Liquid dish washing detergent.
13. Aluminum foil. To cover surfaces to prevent contamination of tools and specimens.
14. Unpainted plywood cutting boards.
15. Pre-baked 4 g amber vials, with caps, for bile samples.
16. IChem prebaked, custody sealed 4 oz jars with Teflon lined caps.

Work Area

The samples should not be taken on a surface painted with oil-based paint. This problem can be solved by laying a sheet of unpainted plywood on top of the painted surface and using aluminum foil to cover work surfaces.

Water dripping from vessel must not be allowed to splash work area.

Preparation of Equipment

At the end of sampling and prior to use, wash all cutting tools, forceps, pans, cutting surfaces, etc. with hot water and detergent. Hot water is available on deck. Rinse thoroughly with hot water and then MC. On cold days the MC may freeze the water. If so, wipe equipment with paper towels prior to rinsing with MC. Flush all tools, pans, etc. with MC and arrange tools for use. Knife blades are positioned so that they do not touch any surface. Scalpels, etc. are kept in the MC rinsed stainless steel pans. Air dry all utensils after rinsing with MC.
Use indelible pen to mark labels, not pencil. NO plastics touch the sample.

Capturing the Specimens

All samples are from internal tissue and thus capturing specimens with otter trawls is valid. Because of large catches, tows are usually limited to 15 minutes. The catch is dumped onto the sorting table and sample specimens immediately put aside for processing.

Selecting the Specimens

Hydrocarbon samples:

Decide beforehand which species and sizes will be sorted from a given catch. Avoid large individuals - their viscera are too large to manipulate effectively. At least four specimens of a given species should be retained from the catch, three for hydrocarbon sampling and one for backup. The selected fish are put in fish baskets and kept cold by covering them with ice.

Histology samples:

Only live or moribund specimens are suitable for processing. Do not process dead fish. Tissues in dead fish autolyze rapidly and mask the subtle changes caused by toxic chemicals. Do not over-ice fish to the extent that tissues freeze. Frozen tissues are worthless for histological examination.

Preparing the Specimen

The specimen is wiped repeatedly with paper towels to remove all slime, mud, scales, etc. The table where the specimen will be placed is also wiped clean.

Taking Samples

Always wear surgical gloves to avoid contamination of samples by

skin oils. Use new gloves rather than trying to reuse gloves. Used gloves are difficult to put on and trying to put them on runs the risk of contamination.

Hydrocarbons:
Removing the pectoral fin creates a hole that allows the knife blade to enter the abdominal cavity without contaminating the internal tissues. Use a short blade boning knife to remove the pectoral fin by slicing the fin away with short strokes in the direction of the head. Wipe knife and put it aside. Remember that each tool must be clean (rinsed with MC) before it touches the sample.

The abdominal wall must be removed to expose the internal organs. Insert the long thin blade of the fillet knife into the pectoral fin hole with the cutting edge of the blade toward the ventral side of the fish, and run the blade very carefully under the surface of the abdominal wall to a point above and behind the anus. The cut is made high enough along the side of the fish that the belly portion prevents the viscera from spilling out onto the work table. The knife point is then pushed through the body wall and the knife pulled downwards and backwards to cut through the flesh. The cut is thus made from the inside out and prevents contamination of the abdominal cavity. The knife is then wiped cleaned and put aside in a clean area.

Subsequent cuts are made holding the body wall away from the internal organs. To lift the body wall, carefully insert the knife point under the cut body wall and lift the body wall up. The body wall is then cut away, the cut being made from the inside out. and the knife wiped and then rinsed with MC before each cut. After the cut, the body flap is discarded, the knife wiped cleaned and put in the stainless steel pan.

Collecting bile

The bile duct opens into the small intestine. The duct is grasped with forceps, gently pulled upwards, and connective tissue around the gall bladder is gently cut away (a deep belly scalpel seems best for this cutting). When free from connective tissue, the gall bladder is set on top of, and slightly to one side of the 4 g amber vial. The #11 straight point scalpel is used to pierce the bladder so that the point of the blade projects inside the vial and acts as a guide for the bile to flow into the vial. Care is needed to prevent the gall bladder or bile from creating a seal or bubble over the top of the vial. Such a seal prevents the bile from flowing into the vial. One technique to aid the puncturing is to hold clean forceps against the bladder on the opposite side from the puncture by the #11 scalpel. Fortunately, only a few drops of bile are needed for analysis. The vial is then capped tightly.

Collecting intestinal tissue

Intestinal tissue includes stomach contents. Indeed, stomach

contents is more valuable for detecting intestinal oil than intestinal tissues themselves. Use forceps (large or small, depending on size of fish) to grasp firmly the esophagus just behind the pharyngeal area. The esophagus is severed anterior to the forceps, and pulled upward as connective tissue is cut away with the deep bellied scalpel. The liver is also cut away. The entire intestine is pulled free of all connective tissue and the lower intestine severed at the anus. Small digestive systems are put directly into the 4 oz IChem jars. Large digestive systems are laid onto a clean and rinsed stainless steel pan and small sections cut from the esophagus, stomach wall, and intestine. These sections and as much of the stomach contents as possible are put into the jar.

Muscle tissue

Muscle tissue is removed from the tail region just posterior to the gut. Using a fillet knife, two longitudinal and parallel cuts are made, one above and the other below the lateral line. Using the same knife, the flesh above and below the two cuts is filletted away. This cutting exposes the sides of the muscle sample such that the knife can be inserted under the sample without contacting the skin and scales. A clean and rinsed knife is used to fillet the sample from the backbone the distance of the two parallel cuts. Care should be taken to avoid contaminating the center area of the sample with the portion of the knife blade that touches the edge of the sample. This care is needed because the edges of the sample could have become contaminated from the fillet knife cutting vertically through the skin.

The rectangular sample is laid skin down on a clean surface. The flesh is shaved away from each side with a scalpel in such a way that the scalpel blade enters the flesh from the clean side and the cuts made so that the blade exits in the unclean areas. The flesh is then filletted from the skin and put into a clean 4 oz IChem jar.

Sampling Crab

Three kinds of tissue are removed from tanner crab (Chionoecetes bairdi), hepatopancreas, muscle, and eggs. A single crab does not provide enough material for a sample and several crab, usually three, are needed to obtain sufficient tissue.

Eggs

The crab are wiped thoroughly with tissue paper. The eggs are removed first. The abdomen is forced apart from the thorax and the outermost (posterior) pair of pleopods excised and discarded. The next two pairs of pleopods with attached eggs are excised and placed into a 4 oz IChem jar. This procedure is repeated on as many crab as needed to provide the sample.

Hepatopancreas

Hepatopancreas is taken from crab of either sex. The carapace is pried off the body of the crab using gloved hands. Part of the hepatopancreas may be lying on the inner surface of the carapace, but most will be in the main body area between the gills. It is a brownish material and is removed with clean forceps directly into a 4 oz IChem jar. As many crab as necessary are use to provide the composite sample.

Muscle

The muscle is removed from the merus segment of each walking leg after the hepatopancreas is removed from the body. First, the merus is severed at its proximal end either by breaking or severing with scissors. To remove the muscle, the joint between the merus and carpus is broken by bending the carpus in the opposite direction that it normally bends. With the joint broken, the carpus is used to pull the muscle out of the merus. The muscle, still attached to the carpus, is held inside the 4 oz jar and the distal end attached to the carpus severed with clean scissors.

Sampling Shrimp

Muscle is taken from the sidestripe shrimp (Pandalopsis dispar). The shrimp is wiped clean, although the interstices between segments is difficult to clean properly. These sections, however, are not likely to come into contact with the muscle sample. Removing shrimp muscle is frustrating, because the muscle tears easily. One procedure is to cut the abdominal exoskeleton laterally along its ventral surface. The abdomen is the spread apart along the cut and held in this position while a second person removes the muscle with clean forceps.

Histology Sampling

Histological sample preparation for fish and shellfish should follow the instructions written by the Histopathology Technical Group.

Chain-of-Custody Procedures

Chain-of-Custody procedures should follow the general instructions "EQM&LO Chain-of-Custody Procedures" (Appendix F). When sample taking must terminate for a short period, such as for meals, a workable procedure to retain custody is to place the on-deck samples in a closed container (such as a Coleman cooler) and seal the container with evidence tape. The tape is broken when sapling is resumed. All sealing and breaking of evidence tape should be noted for record.

APPENDIX F

EQM&LO CHAIN-OF-CUSTODY PROCEDURES

Chain-of-Custody is necessary if there is a possibility that the conclusions based upon analytical data will be used in litigation. The components of chain-of-custody are : sample seals, a field log book, chain-of-custody record, and the Reques for Laboratory Services (RLS); the procedures for their use are described in the following sections.

Due to the evidentiary nature of samples collected during enforcement investigations, possession must be traceable from the time samples are collected until they or their derived data are introduced as evidence in legal proceedings. To maintain and document sample possession, chain-of-custody procedures are followed.

Admissibility of Analyses as Evidence. To be admissible as evidence, samples must be proved conclusively to be in an appropriate person's possession until the analyses resulting therefrom have been introduced as evidence. Rigid controls must be maintained to establish a chain-of-custody for the samples from the time of sampling until ultimate disposition of the particular case.

CUSTODY DEFINITION

A sample is under custody if:

If it is in your possession, or
It is in your view, after being in your possession, or
It was in your possession and you locked it up, or
It is in a designated secure area.

1. Evidence tape or sample seals are used to detect unauthorized tampering of samples following sample collection up to the time of analysis. The seal must be attached in such a way that it is necessary to brea it ~~in order~~ to open the container. Seals must be affixed to containers before the samples leave the custody of sampling personnel.

2. Samples must be kept in such a manner that they cannot be altered wether deliberately or accidentally. Until the samples can be sent to the laboratory they should be kept in a cool, dark, dry place. Refrigeration, freezing or other chemical method of preservation are usually required. Chemical preservatives are added at the laboratory.

   Any indication that a sample has been subjected to tampering or physical alteration could disqualify it a evidence for possible legal action. Therefore, the instructions given herein must be followed strictly.

opening. A evidence tape is placed on the openings of the shipping container, signed and dated.

Sample tags and custody forms must be legible and filled out using waterproof, non-fading ink. Secure individual sample containers or group of sample containers using tamperproof evidence tape or seals.

4. Maintain an up-to-date Field Data Record Logbook. Record field measurements and other pertinent information necessary to refresh the sampler's memory if, later on, he/she takes the stand to testify regarding his/her actions during the evidence gathering activity. Maintain a separate set of field notebooks for each survey; store them in a safe place where they can be protected and accounted for at all times.

5. The field sampler is responsible for the care and custody of the collected samples until they are properly dispatched to the receiving laboratory, or turned over to an assigned custodian. The field sampler should verify that each container is in his/her physical possession or in his/her sight at all times, or is locked so that no one can tamper with it.

6. Colored slides or photographs are often taken to show the outfall sample location and any visible water pollution. Written documentation on the back of the photo should include the photographer's signature, and the time, date and site location. These photographs can be used as evidence, and are handled by chain-of-custody procedures to prevent alteration.

TRANSFER OF CUSTODY AND SHIPMENT

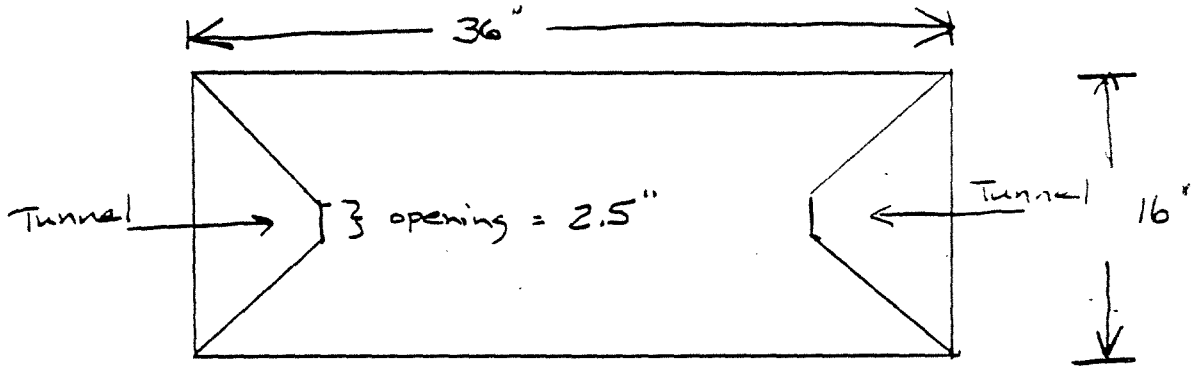
1. Samples are accompanied by a Request for Laboratory Services which has a chain of custody section. When transferring the possession of samples, the individual relinquishing and receiving the samples will sign, date and note the time. This record documents sample custody transfer from the sampler, often through another person, to the laboratory Sample Custodian.

2. Ensure that samples are properly packed in shipping containers (for example, ice chests) to avoid breakage. Ensure that shipping containers are sealed for shipment to the laboratory.

3. If the package is sent by the US mail, ensure that it is sent with a return receipt. If the package is hand-delivered, note that it was hand carried in the method of shipment block in the chain of custody record. Send field receipts from the post office and bills of lading to the laboratory custodian for retention as part of

APPENDIX G.

STANDARDIZED SPOT SHRIMP POT GEAR



½" rebar frame

tunnel webbing = ½" mesh nylon

pot covering = dark nylon fabric

APPENDIX H.

ADF&G SPOT SHRIMP SURVEY
PWS OIL SPILL IMPACT ASSESSMENT PROJECT
FECUNDITY FORM

PROJECT LEADER_____ DATE_____

CRUISE NUMBER_____ SITE_____

STRATUM NUMBER_____ STRING NUMBER_____

SPECIES	CARAPACE LENGTH	TOTAL WET WEIGHT	TOTAL DRY WEIGHT	SUB-SAMPLE WEIGHT	SUB-SAMPLE COUNT

CONFIDENTIAL

STATE/FEDERAL NATURAL RESOURCE DAMAGE ASSESSMENT
DETAILED STUDY PLAN

DRAFT

Project Title: Prince William Sound Oysters

Study ID Number: Fish/Shellfish Study Number 16

Lead Agencies: Alaska Department of Fish & Game
FRED Division
P. O. Box 3-2000; Juneau, Alaska 99802

NOAA/National Marine Fisheries Service
Auke Bay Laboratory
P. O. Box 210155; Auke Bay, Alaska 99821

Principle Investigators: Michael Kaill
Alaska Department of Fish & Game

Malin M. Babcock
NOAA/NMFS Auke Bay Laboratory

STATE OF ALASKA	Signature	Date
Principle Investigator:		10/11/89
Supervisor:		10/11/89
Consulting Biometrician:		10/13/89
OSIAR Senior Biometrician:	N/A	
OSIAR Program Manager:		10/13/89
OSIAR Director:	for	10/17/89

NOAA/NMFS Auke Bay Laboratory

Project Leader: Malin M. Babcock (907) 789-6020		
Damage Assessment Coordinator: Stanley Rice		
Organization Leader: George R. Snyder		10/12/89
Financial Officer: Deborah Rathbone		10/12/89

INTRODUCTION

The goal of this project is to measure the extent to which the Exxon-Valdez oil spill affected oysters in Prince William Sound (PWS). There are no naturally occurring Pacific oysters Crassostrea gigas in Prince William Sound. However, there are three oyster farms, one of which currently has 1.5 million animals on hand. One farm, at Perry Island, is within the oil spill. Another, at Fairmont Island, is near the spill area. The third, Undersea Farms, has oyster racks at two locations - Salmo Point and Deep Bay on the northeast side of Hawkins Island, near Cordova. These latter sites can function as controls (Figure 1). The Exxon Valdez spill may have caused reductions in growth and survival of oysters by direct exposure, by booming or by moving of oysters to substandard habitat to avoid oil contamination.

Oysters have also been used as an indicator species in oil spill impact assessments elsewhere in the world. Similar to the mussel Mytilus edulis, oysters accumulate and bioconcentrate petroleum hydrocarbons (2,000-4,000 x) in their tissues. Oysters probably do not possess the enzyme system necessary to metabolize hydrocarbons; therefore, depuration and return to pre-contamination levels may extend over a long period of time (Blumer et al. 1970; Neff et al. 1985). Petroleum-originating hydrocarbons were found in oysters seven years after the grounding of the Amoco Cadiz (Berthou et al. 1987).

This project will assemble existing information to provide baseline data on growth and survival in oysters. In addition, test stations will be deployed in areas believed to be still exposed to hydrocarbon pollution. This will aid in determining the extent of uptake during the spill, as well as effects of chronic exposure to population growth and survival.

OBJECTIVES

A. Determine the effects of oil contamination on Pacific oyster growth and survival by comparing growth and survival of oysters cultured in oiled and non-oiled environments. The hypotheses will also be tested that the level of hydrocarbons in sediments or mussel tissue is not related to the level of oil contamination of a beach. The experiments are designed to detect a difference of 1.9 standard deviations in hydrocarbon content with the probability of making a type I and type II error of 0.05 and 0.1, respectively.[1]

B. Measure hydrocarbon uptake, depuration and recovery in oysters at all farm site(s) in Prince William Sound.[2]

C. If any loss of production to oyster farms was caused by oil contamination, identify alternative strategies for restoration.[1]

METHODS

This project will consist of three separate activities, each to take place at three sites.

The activities are:

1. Establish an historic data base on growth and survival of oysters for future reference and comparative purposes.

2. Survey hydrocarbon contamination of oysters from all existing oyster farms.

3. Compare oysters in oiled and non-oiled environments to determine:
   a. uptake of hydrocarbons; and,
   b. growth and survival.

Data base

---

[1] This objective will be performed by the Alaska Department of Fish and Game.

[2] This objective will be performed by the National Oceanic and Atmospheric Administration.

At this time, there is no easily accessible long-term data base on growth and survival of oysters in PWS. However, some oyster farmers such as those at Fairmont Island have some historic data available. These data will be obtained and entered into a standard format on R:Base. Historic growth and survival curves will be developed from these data and compared with post oil spill growth and survival data.

Hydrocarbon Surveys

Existing Farms:

This part of the project will establish the extent of injury, and determine rates of recovery from that injury. Oysters were sampled approximately bimonthly at 4 locations in Prince William Sound during April - September 1989 (Table 1).

Whole, unshucked oysters (3 - 10 cm) were placed in precleaned, hydrocarbon-free 16 oz. or 32 oz sample jars or rinsed aluminum foil. Samples were collected to provide at least 10 - 15 g tissue which is the amount necessary for hydrocarbon analyses. Duplicate or triplicate samples were taken. Blanks were taken for the later sampling periods.

Sample jars and lids were purchased precleaned, rinsed with methylene chloride or baked at 440° C for 4 hours. Aluminum foil was rinsed with methylene chloride immediately preceding sampling. Labels included site location, date, time, collector, and species. Latitude and longitude were determined for the locations and this information was included on the Chain-of-Custody Sheets.

All samples were frozen within 1 hour of collection, and samples were transferred to Sid Korn of the Auke Bay Laboratory following rigorous and routine Chain-of-Custody procedures. They are currently being stored frozen at the Auke Bay Laboratory.

Standard Operating Procedures and a Quality Assurance/Quality Control Plan, per se, for this study do not exist. Methods of sample collection follow the general guidelines set forth by the Auke Bay Lab and the Hydrocarbon Technical Committee both in written material and in training sessions.

Growth/Survival Sites:

Triplicate mussel samples sufficient for hydrocarbon analysis (10 - 15 g) will be taken at the beginning and end of the project

from the mean low tide level on the nearest beach to each station (oyster cage). These samples will be analyzed for hydrocarbon content and used as indicators of oil contamination in the vicinity of the cages. Mussels were chosen as an indicator species because of their ability to bioconcentrate hydrocarbons. The mean low tide level will provide a reasonable index of the general location in the water column inhabited by the suspended oysters. Mussel samples will be collected according to the procedure and Chain of Custody outlined above for oysters. Each jar will be labelled with the site name, latitude, longitude, date, "MUSSEL", transect number, sample number, names of the sampling team members, "OYSTER", and "ADF&G".

Three sediment samples for hydrocarbon analyses will be collected from the intertidal area adjacent to each suspended oyster cage at the beginning and end of the project. These sediment samples will provide a representative mixture of sediment composition and contamination from the areas which may be leaching oil to the surface levels inhabited by the suspended oysters. Each sample will be a composite sediment sample which will be collected by scooping approximately 15 cc. sediment to a depth of 2 cm at seven random locations along a vertical transect beginning at the mean low tide level. This will provide a total of 9 samples for the non-oiled impact level (1 hydrocarbon sample/oyster cage * 3 cages/site = 3 hydrocarbon samples/site; 3 hydrocarbon samples/site * 3 sites = 9 hydrocarbon samples/ non-oiled impact level), and 12 samples for the oiled impact level (as above with 4 sites).

Sediment samples from each site will be placed in a 4 oz glass jar rinsed with methylene chloride. Each jar will be labelled with the site name, latitude, longitude, date, "SEDIMENT", transect number, sample number, names of the sampling team members, "OYSTER", and "ADF&G".

The level of sampling described for sediments and mussels is designed to detect differences in petroleum hydrocarbons between impact levels at the desired α and β levels of 0.05 and 0.10, respectively.

Growth/Survival Comparisons

Oyster spat will be deployed in standard test units at four oiled sites (South Perry Island - one site - and Herring Bay at Knight Island - three sites) and three low/non-oiled sites (Deep Bay, Salmo Point, and Fairmont Island). Three test units will be randomly set at each sample site (Figure 2). Each test unit will be a standard 12 mm mesh, 10 layer Japanese lantern net rearing cage (Bourne et al. 1989). It will be anchored 1 m below the surface at low tide and suspended from the bottom with a submerged "trawl" float. A ground line or other means of

locating the unit will be attached to the cage. The cage will be seeded with oyster seed (18 - 20 mm) from the same lot of oyster spat provided by a State of Alaska certified hatchery located outside of the state of Alaska. Seeding rates will be 100 animals per layer for a total of 1,000 animals per cage. One triplicate hydrocarbon sample (three 10 - 15 g samples) will be taken from the initial lot of oyster seed before it is deployed to stations in Prince William Sound waters.

Data and samples will be taken at each site at monthly intervals including: salinity, water transparency (productivity of plant material), number of live and dead oysters, and oyster shell length (the longest dimension out from the hinge) during the growing season (April - October/November). During the winter, samples will only be taken every three months because growth is greatly reduced during winter months (Quayle 1988). At least 30 animals from among the 100 initially placed on each level will be randomly selected from every other layer to be measured. All animals in a layer will be mixed by hand and dumped onto a table that is marked into three trisects. All animals in one of the trisects will be measured. If thirty animals are not measured, the additional animals needed will be randomly selected from the other trisects. Layers to be measured will be alternated during each sampling period (ie. during the first sample period oysters in layers 1, 3, 5, 7, and 9 will be measured and during the second period oysters in layers 2, 4, 6, 8, and 10 will be measured). Sampling five levels per cage will allow 150 specimens (30 specimens X 5 levels) to be measured in each cage, or 450 at each site (150 specimens X 3 cages). This will provide an adequate number of oysters for growth analysis by site and treatment level.

The number of live and dead oysters in each sampled level will also be counted. All available dead shells will be measured to record approximate time of death. After being measured, all dead oysters will be discarded. All data will recorded on a field data form (Appendix A). Measurements will be made with a digital caliper, which allows direct input to the memory of a portable micro computer. Temperature will be recorded automatically with a solid-state temperature recorder (Ryan tempmentor brand) fastened to the cage at layer five.

Twice a year a composite sample of 15 g of oyster tissue will be taken to evaluate hydrocarbon content. The number of oysters to be included in the sample will depend on the size of the animals at the time of sampling. Since all oysters began as spat of generally uniform size, the oysters in each cage are expected to grow at a uniform rate and do not need to be sampled according to a size criterion. Oysters will be randomly selected from all layers to make up each hydrocarbon sample. Hydrocarbon sampling and analyses will be as described previously.

DATA ANALYSES

Hydrocarbon Surveys

Existing Farms:

Parametric statistics (analysis of variance) will be used to test for significant differences ($p \leq .05$) between sites and also levels of tissue hydrocarbons over time. In cases where use of parametric statistics is not possible, nonparametric statistical methods will be used to ascertain differences between sites. Pairwise comparisons (Tukey or Student's t) can also be used for this purpose.

Growth/Survival Sites:

Analysis of variance will be used to test for differences in hydrocarbon levels in both sediments and mussel tissue between sites.

Growth/Survival Comparisons

To examine oyster survival, the proportion of dead oysters to live oysters will be subjected to analysis of variance by site and by oil impact levels.

Differences in incremental growth between treatment levels will be compared with any historic data on oyster growth in Prince William Sound. Growth parameters will be determined for various growth curves, such as Gompertz, von Bertalanffy or polynomial equations. Growth parameters will be presented for the most appropriate growth models only. An analysis of variance on growth parameters obtained from fitting algorithms for oyster growth during the study will be compared with growth parameters obtained from historical data. Analysis of variance will also be used to compare the growth parameters obtained at each treatment level to detect differences in growth parameters due to oil contamination level. Graphics will be used to display differences in growth among areas including length at age for each site.

SCHEDULES AND REPORTS

Dates	Activity
April 1989 to October 1989	Oysters were sampled on an approximate bimonthly schedule to establish hydrocarbon uptake and accumulation, and depuration at existing farm sites.
October 1989 to February 1990	Project staging: sample sites selected, test cages assembled and deployed (October), monthly sampling initiated, data base format initiated and data processing initiated. Preliminary report completed by December 21.
May 1990 to October 1990	Active field season. Based on preliminary report, continuation of data base work to allow comparison of "normal" growth and survival to that of oiled populations.
	Continued sampling of oysters exposed to oil spill to establish depuration rates.

Schedules for report writing and detailed plans set by the Trustee Managers will be met.

PROJECT BUDGET

ADF&G[1]

Line Item	Category	Budget
100	Personnel Services	$ 8,300
200	Travel	$ 5,700
300	Contractual	$ 5,300
400	Commodities	$ 2,000
500	Equipment	$ 2,700
700	Grants	$ 0
Total		$ 25,300

[1] Budget is for all activities performed from March 27, 1989 to February 28, 1990.

FUNDED PERSONNEL

Class	PCN	Name	PFT_mm	SFT_mm
FB II				2.0

NOAA Auke Bay Laboratory

The budget for NOAA/NMFS/Auke Bay Laboratory, for the period 13 April 1989 to 28 February 1990:

Labor	3620	(3/4 month for M. Babcock)
Travel		
Contracts	880	
Supplies	500	
Equipment		

TOTAL $	5000	

LITERATURE CITED

Berthou, F., G. Balouet, G. Bodennec and M. Marchand. 1987. The occurrence of hydrocarbons and histopathological abnormalities in oyster for seven years following the wreck of the Amoco Cadiz in Brittany (France). Mar. Environ. Res. 23:103-133.

Bourne, N., C. A. Hodgson, and J. C. Whyte. 1989. A manual for scallop culture in British Columbia. Canadian Technical Report of Fisheries and Aquatic Sciences No. 1694. 215 pp.

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Neff, Jerry M., Paul D. Boehm and William E. Haensly. 1985. Petroleum contamination and biochemical alterations in oysters (Crassostrea gigas) and plaice (Pleuronectes platessa) from bays impacted by the Amoco Cadiz crude oil spill. Mar. Environ. Res. 17:281-283.

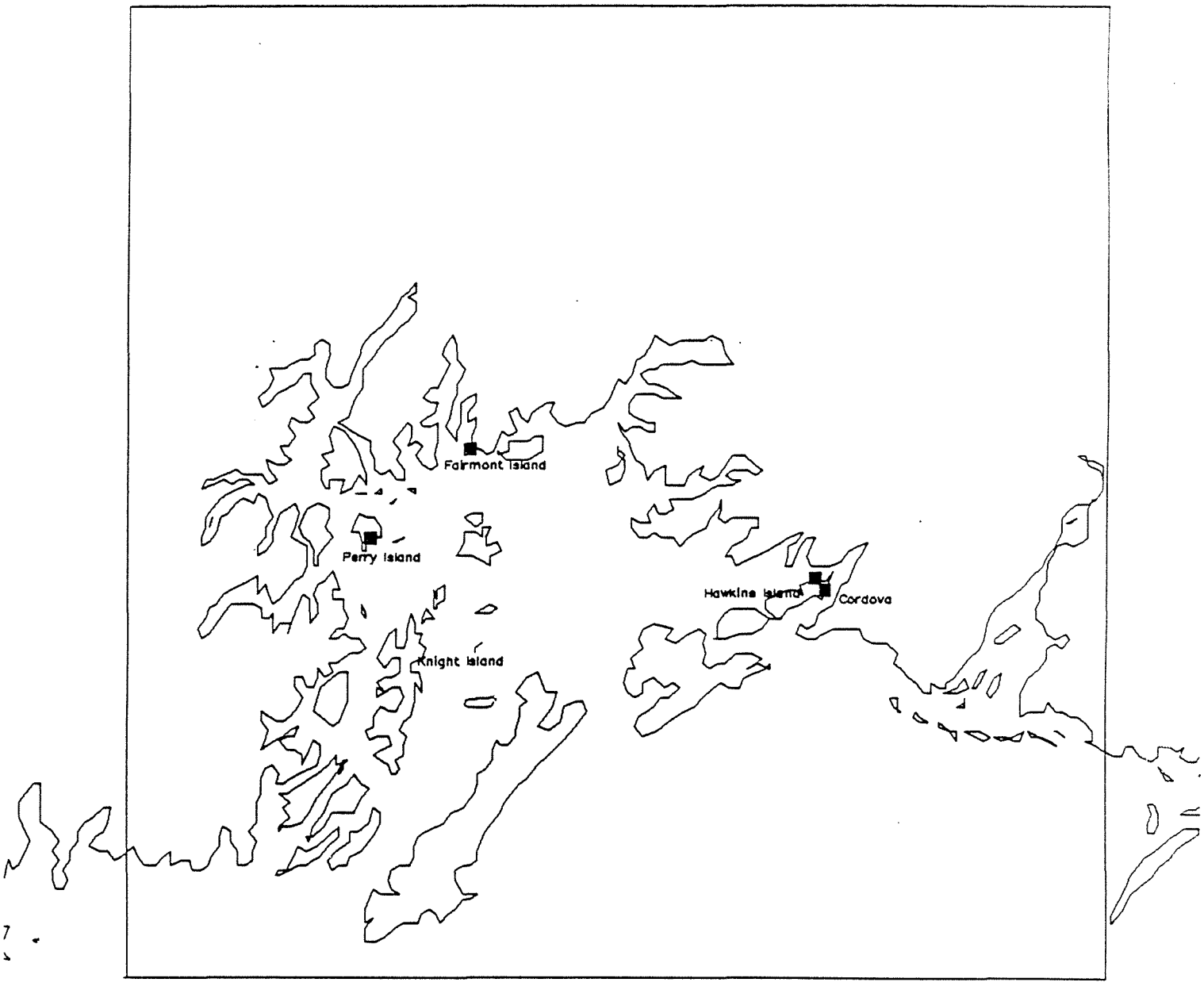
Quayle, D. B. 1988. Pacific oyster culture in British Columbia. Canadian Bulletin of Fisheries and Aquatic Sciences No. 218. 241 pp.

Stegeman, J. J. and J. M. Teal. 1973. Accumulation, release and retention of petroleum hydrocarbons by the oyster Crassostrea virginica. Mar. Biol. 22:37-44.

Table 1. Summary of oyster samples collected from Prince William Sound, 1989, following the EXXON VALDEZ spill. Samples collected by NOAA National Marine Fisheries Service.

BATCH	ID	COLLECTOR	LAT.	LONG.	SITE	WHAT	TYPE	DATE
V89_08	1391	CLARK	60.35.12.	145.46.56.	DEEPBAY	OYST	WHOLE	04/14/89
V89_08	1392	CLARK	60.35.12.	145.46.56.	DEEPBAY	OYST	WHOLE	04/14/89
V89_27	2723	MOLES	60.35.12.	145.46.56.	DEEPBAY	OYST	WHOLE	05/17/89
V89_42	4515	BABCOCK	60.35.12.	145.46.56.	DEEPBAY	OYST	WHOLE	06/21/89
V89_42	4516	BABCOCK	60.35.12.	145.46.56.	DEEPBAY	OYST	WHOLE	06/21/89
V89_72	8286	FLETCHER	60.35.12.	145.46.56.	DEEPBAY	-0-	-0-	09/19/89
V89_72	8284	FLETCHER	60.35.12.	145.46.56.	DEEPBAY	OYST	WHOLE	09/19/89
V89_72	8285	FLETCHER	60.35.12.	145.46.56.	DEEPBAY	OYST	WHOLE	09/19/89
V89_72	8283	FLETCHER	60.35.12.	145.46.56.	DEEPBAY	OYST	WHOLE	09/19/89
V89_08	1395	CLARK	60.53.36.	147.26.45.	FAIRMONT	OYST	WHOLE	04/13/89
V89_08	1396	CLARK	60.53.36.	147.26.45.	FAIRMONT	OYST	WHOLE	04/13/89
V89_27	2724	MOLES	60.53.36.	147.26.45.	FAIRMONT	OYST	WHOLE	05/13/89
V89_70	7982	O'CLAIR	60.53.36.	147.26.45.	FAIRMONT	OYST	WHOLE	09/11/89
V89_70	7984	O'CLAIR	60.53.36.	147.26.45.	FAIRMONT	OYST	WHOLE	09/11/89
V89_08	1393	CLARK	60.40.55.	147.55. 0.	PERRY	OYST	WHOLE	04/13/89
V89_08	1394	CLARK	60.40.55.	147.55. 0.	PERRY	OYST	WHOLE	04/13/89
V89_27	2725	MOLES	60.40.55.	147.55. 0.	PERRY	OYST	WHOLE	05/13/89
V89_	5833	RICE	60.40.55.	147.55. 0.	PERRY	OYST	WHOLE	07/12/89
V89_70	7978	O'CLAIR	60.40.55.	147.55. 0.	PERRY	OYST	WHOLE	09/10/89
V89_70	7980	O'CLAIR	60.40.55.	147.55. 0.	PERRY	OYST	WHOLE	09/10/89
V89_70	7981	O'CLAIR	60.40.55.	147.55. 0.	PERRY	-0-	-0-	09/10/89
V89_70	7979	O'CLAIR	60.40.55.	147.55. 0.	PERRY	OYST	WHOLE	09/10/89
V89_08	1397	CLARK	60.35.38.	145.48. 0.	SALMOPT	OYST	WHOLE	04/14/89
V89_08	1398	CLARK	60.35.38.	145.48. 0.	SALMOPT	OYST	WHOLE	04/14/89
V89_27	2722	MOLES	60.35.38.	145.48. 0.	SALMOPT	OYST	WHOLE	05/17/89
V89_42	4513	BABCOCK	60.35.38.	145.48. 0.	SALMOPT	OYST	WHOLE	06/21/89
V89_42	4514	BABCOCK	60.35.38.	145.48. 0.	SALMOPT	OYST	WHOLE	06/21/89
V89_72	8280	FLETCHER	60.35.38.	145.48. 0.	SALMOPT	OYST	WHOLE	09/19/89
V89_72	8281	FLETCHER	60.35.38.	145.48. 0.	SALMOPT	OYST	WHOLE	09/19/89
V89_72	8282	FLETCHER	60.35.38.	145.48. 0.	SALMOPT	OYST	WHOLE	09/19/89

Figure 1. - Oyster sampling sites in Prince William Sound



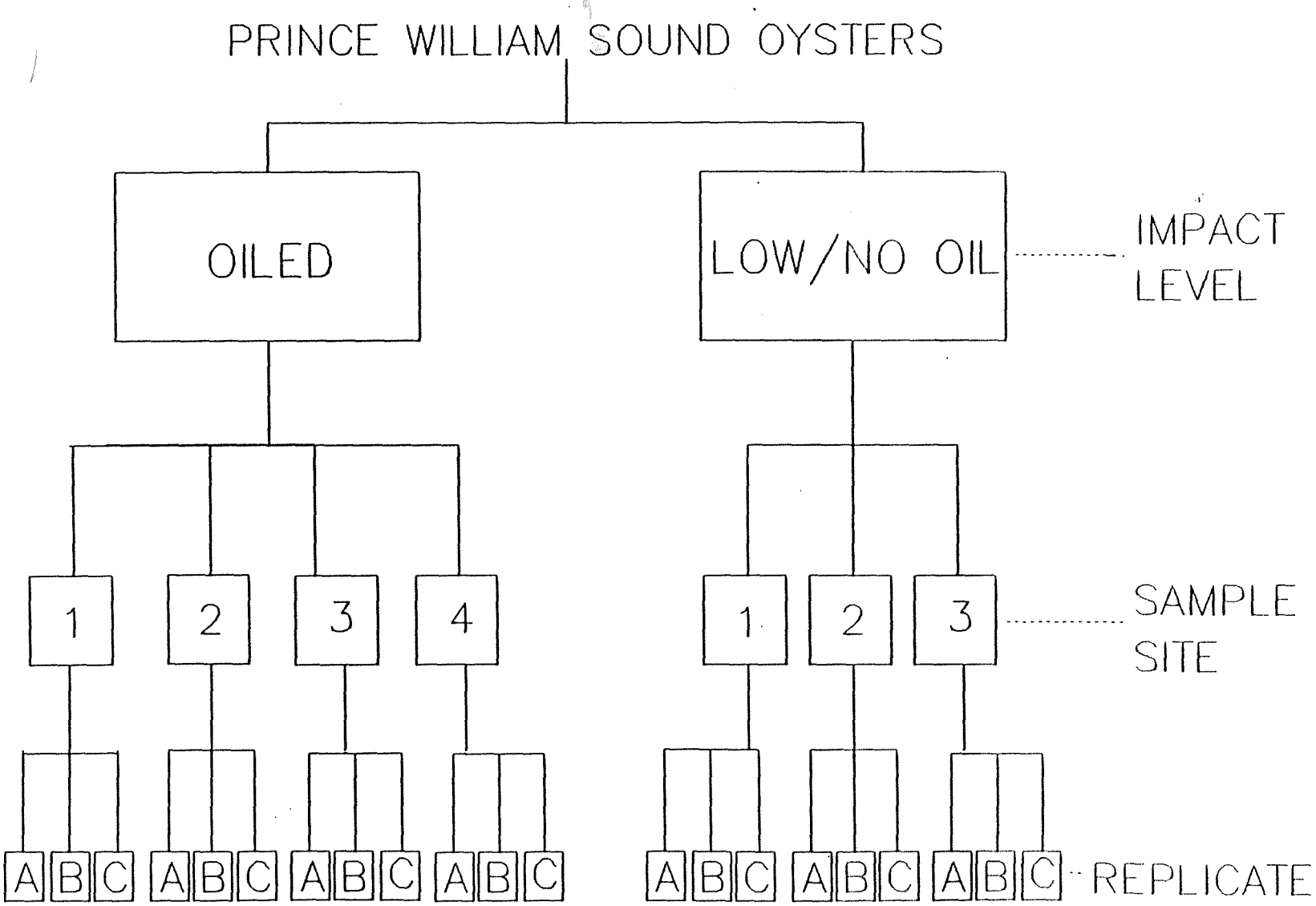


Figure 2. Sampling desing for growth and survival comparisons of oysters in Prince William Sound.

Appendix A. Field data form.

ADF&G PRINCE WILLIAM SOUND OYSTER PROJECT

DATE_____ SITE_____

SUBSITE_____ CAGE NUMBER_____

WATER TEMPERATURE_____ (degrees C) WATER SALINITY_____0/00

WATER TRANSPARENCY_____ (meters) WATER COLOR_____

LAYER NUMBER_____ RECORDER NAME_____

NUMBER OF LIVE OYSTERS_____

NUMBER OF DEAD OYSTERS (0 - 20 mm.)_____

NUMBER OF DEAD OYSTERS (21 - 30 mm.)_____

NUMBER OF DEAD OYSTERS (31 - 40 mm.)_____

LENGTH FREQUENCY

OYSTER NUMBER	LENGTH	OYSTER NUMBER	LENGTH	OYSTER NUMBER	LENGTH

STATE/FEDERAL NATURAL RESOURCE DAMAGE ASSESSMENT
DETAILED STUDY PLAN

Project Title: Prince William Sound Oysters

Study ID Number: Fish/Shellfish Study Number 16

Lead Agencies: Alaska Department of Fish & Game
FRED Division
P. O. Box 3-2000; Juneau, Alaska 99802

NOAA/National Marine Fisheries Service
Auke Bay Laboratory
P. O. Box 210155; Auke Bay, Alaska 99821

Principle Investigators: Michael Kaill
Alaska Department of Fish & Game

Malin M. Babcock
NOAA/NMFS Auke Bay Laboratory

STATE OF ALASKA	Signature	Date
Principle Investigator:	_____	_____
Supervisor:	_____	_____
Consulting Biometrician:	_____	_____
OSIAR Senior Biometrician:	_____	_____
OSIAR Program Manager:	_____	_____
OSIAR Director:	_____	_____

NOAA/NMFS Auke Bay Laboratory

Project Leader: Malin M. Babcock (907) 789-6020	_____	_____
Damage Assessment Coordinator: Stanley Rice	_____	_____
Organization Leader: George R. Snyder	_____	_____
Financial Officer: Deborah Rathbone	_____	_____

CONFIDENTIAL

STATE/FEDERAL RESOURCE DAMAGE ASSESSMENT
DETAILED STUDY PLAN

Project Title:	INJURY TO ROCKFISH IN PRINCE WILLIAM SOUND
Study ID Number:	Fish/Shellfish Study Number 17
Lead Agency:	State of Alaska, ADF&G; Sport Fish Division
Cooperating Agencies:	Federal: USFS State: DNR
Principal Investigator:	Kelly Hepler, Fishery Biologist
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INTRODUCTION

The goal of this project is to determine whether the Exxon-Valdez oil spill will have a measurable impact on stocks of rockfish, *Sebastes sp.*, in Prince William Sound (PWS). Assemblages of nearshore fish populations of PWS support low volume commercial and recreational fisheries. Most of the annual harvest is supported by only a few species of rockfish, although several other species are harvested in small numbers. Of particular importance are: yelloweye rockfish *S. ruberrimus*; dusky rockfish *S. ciliatus*; black rockfish *S. melanops*; and quillback rockfish *S. maliger*. Unlike many species of marine fish, demersal rockfish complexes are relatively sedentary, residing near rocky reefs and boulder fields. Rockfish are long-lived, recruitment is low, and the potential for long-term stock decline due to habitat degradation is high. The potential impact of the oil spill on various nearshore assemblages is dependent upon location of various "rockpiles" and the potential uptake of various contaminants will be related to the level of oil contamination and food web characteristics of these various reefs.

Data Base

Only limited baseline data are available for rockfish populations in PWS. Rockfish were studied as part of a study of nearshore fish assemblages during the years 1977-1979 (Rosenthal 1980). These investigations provided descriptions of selected rockfish populations including estimates of species composition, density, and length and age composition. This sampling primarily occurred at four major sampling sites.

Experimental Design

It is hypothesized that several detrimental impacts on these species could result from the presence of crude oil in marine waters including: (1) reduced survival; (2) reduction in reproductive success; and (3) accumulation of sublethal levels of toxic petrochemical by-products that could render the fish inedible. To test whether there will be a measurable impact on these stocks, selected locations in each of two treatments will be sampled: oiled and non-oiled areas.

The principal objectives of the project are to document presence/absence of: (1) rockfish in areas where rockfish are known to have previously occurred; and (2) oil contamination in rockfish and/or the substrate. Our primary assumption is that there is a difference in exposure to oil for fish stocks from each of the two treatments. Evidence from the literature indicates that the greatest observable impact of oil related activities occur in the littoral zone (National Academy of Sciences 1985); an area which supports populations of rockfish.

A measurable detrimental impact on these stocks of rockfish may result in a loss to the sport fishery. The status of the sport fishery will be investigated through: (1) an ongoing postal survey (Mills 1988) and (2)

an onsite creel survey of selected PWS fishery access ports (OSIAR Study FS #6).

OBJECTIVES

1. Document the presence or absence of rockfish in 13 locations in Prince William Sound.

2. Document the presence or absence of oiled rockfish in 13 locations in Prince William Sound.

3. Document the presence or absence of oiled substrate in 13 locations in Prince William Sound.

4. Identify potential alternative methods and strategies for restoration of lost use, populations, or habitat where injury is identified (to be accomplished upon completion of the project).

METHODS

This operational plan addresses both the long range study design of this project and work that has already been accomplished as part of the initial fishery impact assessment work. Throughout the remainder of this document, work that has already been initiated will be identified. Because of the immediacy of the initial fishery impact assessment work, some of the experimental design had not been fully developed and certain aspects of the sampling were not initiated. Changes in sampling design will be identified for initiation in the future.

Study Design and Data Collection

Thirteen sites will be sampled across two treatments: oiled and non-oiled areas (Table 1). Four of these sample sites correspond to areas which have been previously studied during the 1977-1979 OCSEP survey (Rosenthal 1980). During the OCSEP survey, transects were established and sampled for density by species information. This information will provide the basis for determining the historic presence or absence of rockfish. The remaining nine sample sites were selected after consulting with local fisherman and from areas of reported fish kills. Sampling will be conducted during at least two surveys over the course of the summer. The first survey occurred within five weeks of the spill. The second survey occurred during August, which corresponds to the sampling period of peak abundance in Rosenthal's work (1980).

At each location, transects will be established and sampling will be conducted along the majority of the reef with longline gear. The exact location of each transect will be identified with LORAN and/or latitude/longitude coordinates at each end of the transect. In addition to coordinates, depth along the transect and a compass reading from the starting to ending point will be recorded. If fish are present, species

composition will be estimated for comparison with Rosenthal's dive surveys (1980). In addition, samples of more pelagic species will be collected by jigging along the face of the reef. Species identification of rockfish will be accomplished using the methods of Kramer and O'Connell (1988).

Sampling for oil contamination will be accomplished as follows. Ten rockfish from each sampling location will be collected for hydrocarbon testing. Each entire fish will be wrapped in aluminum foil, marked with an evidence seal, and frozen. Upon return to port, the following samples will be collected for hydrocarbon analysis: gallbladder, stomach, pyloric caeca, liver, and muscle. Procedures to prevent hydrocarbon contamination, as specified by the National Marine Fisheries Service Auke Bay Lab, will be followed for each fish: (1) hands and sampling gear will be washed with soap and water; (2) dissection tools will be rinsed in methylene chloride; (3) samples of each tissue will be individually stored in certified hydrocarbon-free sampling jars; and (4) samples will be frozen. Samples will not be touched by human hands or any petrochemical product (i.e. plastic). These samples will be transferred to National Marine Fishery Service for analysis. Care must be taken to follow chain of custody procedures. In addition samples will be collected for organoleptic testing. Samples will be handled as described above except that the samples will be iced instead of frozen and the samples will be transferred to Alaska Department of Environmental Conservation for analysis. Additionally, any moribund fish that are found will be examined for presence of tar balls in the stomach and all contaminated samples will be handled in the prescribed chain of custody procedures.

Sampling has already been conducted as part of the initial fishery impact assessment work. Sampling was initiated during the first week of May and mid-June. Hydrocarbon sampling during the mid-June survey was conducted in only three locations. Sampling was again conducted during mid-September, the time frame that Rosenthal (1980) identified as near the peak abundance of rockfish in nearshore areas.

The presence/absence of oil at each sampling site will be documented with a Remote Operating Vehicle (ROV). This work is covered under another project (Undersea Observations to Support Benthic Fishery Damage Assessment). Visual (video tape) records will be collected along transects within rockfish sampling areas. Presence of oil, general distribution, occurrence of rockfish, depth, substrate type, turbidity, temperature, and salinity will be recorded. Transect density will be increased where evidence of oil is found. If oil is observed, subsequent sampling will be initiated to collect a sample for verification. ROV work was only accomplished during the mid-June survey.

Data Analysis

Data analysis will be limited to a simple expression of presence or absence; percent of sample contaminated; and degree of contamination (ppm of hydrocarbons). Proportions of all nonoiled sites with rockfish

will be compared to the proportions of all oiled sites with rockfish on a species by species basis. Hydrocarbon contamination will be detected 99% ($\alpha = .01$) of the time if over 37% of the population is contaminated at the selected sample size of 10.

SCHEDULES

A schedule of tasks to be completed during 1989 is as follows:

Task	Dates
Sampling: 1st Survey	Early May
2nd Survey	Late June
3rd Survey	Mid-September
Data Analysis and Report Preparation	9/15-3/15

REPORTS

Results of these study efforts will be reported to the Division of Oil Spill Impact Assessment and Restoration. Upon completion of litigation, these data will be published as either an Alaska Department of Fish and Game, Sport Fish Division, Fishery Data Series report or in the fisheries literature.

BUDGET SUMMARY

A line item breakdown of project costs for the period beginning April 1, 1989 and ending February 28, 1990 is as follows:

Line Item	Category	Cost (thousand $)
100	Personnel	12.0
200	Travel	3.6
300	Services	23.0
400	Commodities	6.0
500	Equipment	1.0
	Total	45.6

LITERATURE CITED

Kramer, D.E. and V.M. O'Connell. 1988. Guide to Northeast Pacific Rockfishes Genera Sebastes and Sebastolobus. University of Alaska Marine Advisory Bulletin No. 25.

Mills, M.J. 1988. Alaska statewide sport fisheries harvest report. Alaska Department of Fish and Game, Fishery Data Series No. 2.

Rosenthal, R.J. 1980. Shallow water fish assemblages in northeastern gulf of Alaska: habitat evaluation, species composition, abundance, spatial distribution and trophic interaction. Prepared for NOAA, OCSEAP Program. 84 pp.

National Academy of Sciences 1985. Oil in the Sea: Impacts, Fates, and Effects. National Academy Press, Washington DC. 601 pp.

Table 1. Sampling locations for rockfish in PWS, 1989.

Location		Longitude	Latitude	LORAN
Windy Bay	Start	145° 58.29′	60° 34.33′	14325.0/31948.7
	End	145° 59.07′	60° 34.46′	14324.1/31946.0
Knowles Head	Start			
	End	146° 33.95′	60° 38.73′	
Bligh Reef[1]		146° 52.75′	60° 51.20′	
Naked Island[1]	Start	147° 28.78′	60° 42.11′	
	End	147° 29.36′	60° 42.15′	
Northwest Bay[1]	Start	147° 37.29′	60° 34.02′	
	End	147° 37.68′	60° 33.98′	
Herring Bay[1]	Start			13889.5/32035.5
	End			13886.9/32033.4
Pt. Nowell[1]	Start	147° 55.60′	60° 25.99′	
	End	147° 56.08′	60° 25.41′	
Applegate Is.[1]	Start	148° 08.53′	60° 36.59′	
	End	148° 07.49′	60° 36.32′	
N Danger Is.[1]	Start			13489.6/31814.6
	End			13485.7/31813.5
S Danger Is.[1,2]				13473.0/31808.0
Schooner Rock[2]	Start	146° 03.73′	59° 43.08′	
	End	146° 05.25′	59° 43.16′	
Zaikoff Bay[2]	Start	146° 56.51′	60° 18.77′	
	End	146° 57.16′	60° 18.73′	
Port Etches[2]	Start			14026.1/31880.4
	End			14023.9/31878.5
Porpoise Rks.				14024.0/31881.0

[1] Oiled sites
[2] Sites sampled by Rosenthal (1980)