# **Operational Plan: Northern Cook Inlet Chinook Salmon Marine Harvest Composition Study**

by Adam St. Saviour, Chase Jalbert, Bronwyn Jones, and Andrew W. Barclay

January 2015

Alaska Department of Fish and Game

**Divisions of Sport Fish and Commercial Fisheries** 



#### Symbols and Abbreviations

The following symbols and abbreviations, and others approved for the Système International d'Unités (SI), are used without definition in the following reports by the Divisions of Sport Fish and of Commercial Fisheries: Fishery Manuscripts, Fishery Data Series Reports, Fishery Management Reports, and Special Publications. All others, including deviations from definitions listed below, are noted in the text at first mention, as well as in the titles or footnotes of tables, and in figure or figure captions.

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## **REGIONAL OPERATIONAL PLAN SF.2A.2015.01**

### OPERATIONAL PLAN: NORTHERN COOK INLET CHINOOK SALMON MARINE HARVEST COMPOSITION STUDY

by

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January 2015

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### SIGNATURE/TITLE PAGE

Project Title:	Northern Cook Inlet Chinook Salmon Marine Harvest
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Division, Region, and Area	Sport Fish Division, Region II, Anchorage
Project Nomenclature:	
Period Covered:	May 2015 - January 2016
Field Dates:	May 15 - September 15, 2015
Plan Type:	Category III

## Approval

Title	Name	Date
Project Leader	Adam St. Saviour	1/50/ZOIS
Co-Project Leader	Andrew Banday	1/30/15
Biometrician	Azton Antonovich	2/2/15
Regional Research Supervisor	Tim McKinley	3/3/11
Fish and Game Coordinator	Most Muse	2/2/15

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## ABSTRACT

Chinook salmon (*Oncorhynchus tshawytscha*) runs have been historically low throughout southcentral Alaska in recent years. Chinook salmon are harvested in mixed stock fisheries of the Northern District of Upper Cook Inlet (UCI). There are eight Chinook salmon stocks designated as stocks of concern by the Alaska Board of Fisheries that must pass through the Northern District of Cook Inlet immediately before reaching their natal streams. The community of Tyonek is reliant on subsistence harvest of Chinook salmon on the west side of the Northern District. Chinook salmon harvested in the Tyonek subsistence and UCI Northern District set gillnet commercial fisheries will be sampled at ports, processors, and on the fishing grounds for genetic tissue, scales, sex, length, maturity, and coded wire tags. Genetic mixed stock analysis techniques will be used to estimate the proportion and number of Chinook salmon harvested in these fisheries by reporting group (*UCI Northwest, Susitna-Matanuska, Knik-Turnagain, and Kenai Peninsula*) for temporal and geographic strata. Age, sex, length, and maturity compositions will be estimated in the Tyonek subsistence and UCI Northern District set gillnet commercial fisheries overall.

Keywords: Chinook salmon, mixed stock analysis, Northern District of Upper Cook Inlet, set gillnet, subsistence, Tyonek

### PURPOSE

The primary purpose of the Chinook salmon marine harvest project is to estimate the stockspecific harvests of Chinook salmon from the Tyonek subsistence fishery and the Northern District commercial set gillnet fishery to improve understanding of stock productivity. Stockspecific harvests will be estimated by collecting and analyzing genetic tissue samples from fish caught in these fisheries. This information will help inform the calculations for escapement goals and will assist in the management of the Chinook salmon runs that support the Tyonek subsistence and Northern District commercial set gillnet fisheries.

### BACKGROUND

All five species of Pacific salmon are harvested in upper Cook Inlet (UCI). Sockeye salmon (*Oncorhynchus nerka*) make up the majority of the harvest (Shields and Dupuis 2013b) but Chinook salmon (*O. tshawytscha*) are also harvested. Chinook salmon returning to the Susitna River and west Cook Inlet streams are harvested in the mixed stock marine fisheries of UCI, primarily the Tyonek Subdistrict subsistence set gillnet and the Northern District commercial set gillnet fisheries (Figure 1). Recent low Chinook salmon runs throughout Southcentral Alaska, especially to UCI, have heightened concerns about stock-specific harvest of Chinook salmon in these fisheries. There are now eight Chinook salmon stocks in the Northern District of Cook Inlet that are designated as stocks of concern by the Alaska Board of Fisheries (BOF; Alaska Department of Fish and Game; ADF&G, 2014). Three of these stocks of concern (Theodore, Lewis, and Chuitna rivers) likely comprise the majority of the Chinook salmon harvested in the Tyonek Subdistrict subsistence fishery. These three streams are the most productive Chinook salmon rivers flowing into the West Cook Inlet management area and are adjacent to the community of Tyonek.

However, little, if any, information regarding stock-specific harvests of Chinook salmon in these mixed-stock marine fisheries is currently available. Tyonek residents have stated that they believe most Chinook salmon harvested in the subsistence fishery in marine waters are returning to the Chuitna River; however, it is likely a portion of Chinook salmon harvested in the subsistence fishery are returning to the Susitna River drainage.

A Chinook salmon genetic baseline that includes representative Pacific-wide populations is now available for mixed stock analysis (MSA) applications (Templin et al. 2011). An additional

Chinook salmon genetic baseline that includes extensive sampling of populations within Cook Inlet is nearing completion (Barclay et al. *In prep*). This unpublished baseline has been successfully used in a similar MSA study of Chinook salmon harvested in the Upper Subdistrict of the Central District set gillnet fishery, also known as the "East Side Set Net" fishery. This project will sample Chinook salmon from the Tyonek subsistence set gillnet and Northern District commercial set gillnet fisheries for three years. The unpublished Cook Inlet baseline will be used to estimate the stock-specific harvests.

The first year of sampling occurred in 2014. During this first season, staff port-sampled commercially-caught Chinook salmon on six Northern District set gillnet openers (June 2, 9, 16, 23, 26, and 30). Chinook salmon were sampled for genetics and usually for age, sex, and length. Crews were simultaneously deployed to two processors and one boat ramp in Anchorage, one to three processors or pick up locations around Nikiski, and on the fishing grounds in Tyonek. Approximately 774 Chinook salmon were sampled from the commercial fishery of a reported harvest of 1,473. The percentage of reported catch that was sampled ranged from 17 to 100% for all openers and locations. The season average was 52.5%. The manner in which Chinook salmon were brought in and sold from this fishery was highly variable each week. It required a lot of good communication with processors and samplers to maximize the number of Chinook salmon sampled.

Subsistence Division sampled the subsistence openers in Tyonek. Subsistence fishing was open Tuesdays, Thursdays, and Fridays May 15 through June 15 and then just Saturdays from June 16 on. Subsistence Division sampled all openers through June 28 with the exceptions of 5/20-27 (due to fire) and 6/13 (for a funeral).

A total of 202 Chinook salmon genetic and age/sex/length (ASL) samples were collected from the Tyonek subsistence fishery along with ethnographic data. Total 2014 subsistence harvest reporting was not complete at time of publication.

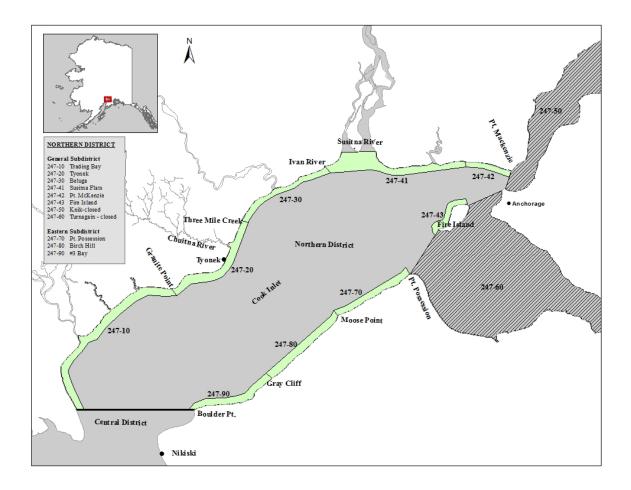


Figure 1.–Map of Upper Cook Inlet Northern District set gillnet commercial fishing statistical areas.

## **OBJECTIVES**

- 1. Estimate the proportion of Chinook salmon harvested in the Tyonek subsistence and UCI Northern District set gillnet commercial fisheries by reporting group (UCI Northwest, Susitna-Matanuska, Knik-Turnagain, and Kenai Peninsula) for each temporal and geographic stratum such that the estimated proportions are within 22 percentage points of the true values 90% of the time.
- 2. Estimate the age composition of the Chinook salmon harvested by the Tyonek subsistence and UCI Northern District set gillnet commercial fisheries such that the estimates are within 10 percentage points of the true values 95% of the time.

#### **SECONDARY OBJECTIVES**

- 1. Sample 70% of the Chinook salmon harvested in the Tyonek subsistence and UCI Northern District set gillnet commercial fisheries for tissue, coded-wire tags (CWTs), scales, sex, and mid-eye- fork (MEF) length.
- 2. Estimate the harvest of Chinook salmon in the Tyonek subsistence and UCI Northern District set gillnet commercial fisheries by reporting group (UCI Northwest, Susitna-Matanuska, Knik-Turnagain, and Kenai Peninsula).

3. Estimate the sex, length, and maturity compositions of Chinook salmon harvested in the Tyonek subsistence and UCI Northern District set gillnet commercial fisheries overall such that the estimates are within 10 percentage points of the true values 95% of the time.

## **METHODS**

#### **GEOGRAPHIC AND TEMPORAL STRATIFICATION**

Proposed temporal and geographic stratification for the UCI Northern District set gillnet commercial fishery was determined based on location and reported harvest data from 2011-2014 (Shields and Dupuis 2011, 2013a, 2013b, *In prep*). Two temporal and three geographic strata were chosen (Table 1).

Stratum #	Temporal Stratum	Geographic Area
1	25 May – 15 June	Trading Bay
2		Pt. McKenzie, Susitna Flats, and Fire Island
3		Eastern Sub-district
4	16 - 30 June	Trading Bay
5		Pt. McKenzie, Susitna Flats, and Fire Island
6		Eastern Sub-district

Table 1.–Proposed strata for ASL and MSA in 2015.

The 2011-2014 reported harvests were used to determine expected harvests and also expected sampling rates for 2015 (Table 2). As in-season sampling to represent to harvest is not possible, subsampling of collections is performed postseason to ensure equivalent representation of the harvest. The goal for 2015 will be to collect tissue from 70% of the harvest. Based on the average 2011-2014 harvests, 70% sampling rate should provide at least 100 samples from most of the selected strata.

Table 2.–Reported Chinook salmon harvest by temporal and geographic stratum in the UCI Northern District set gillnet fishery, 2011-2014.

Temporal	Geographic Area		2011-2014			
Stratum		2011	2012	2013	2014	Average
28 May – 15 June	Trading Bay	423	164	296	384	317
	Pt. McKenzie, Susitna Flats, and Fire Island	475	120	394	287	319
	Eastern SubDistrict	198	95	146	219	165
16 - 30 June	Trading Bay	159	262	165	192	195
	Pt. McKenzie, Susitna Flats, and Fire Island	233	98	60	130	130
	Eastern SubDistrict	131	109	73	93	102

To analyze samples collected from the Tyonek subsistence fishery, we will use two-week temporal strata. However, if it is determined that the sampling has been done proportional to the harvest, then no stratification will be necessary.

The objective criteria ( $\pm$  0.10 with 95% confidence level) for estimating the age composition of Chinook salmon harvested in the Tyonek subsistence and UCI Northern District set gillnet commercial fisheries should be achieved with approximately 170 scale samples. To arrive at this sample size we assumed a worst-case scenario of 25% scale regeneration rate with multinomial proportions of equality among ages (Thompson 1987). As we plan to collect more samples in 2015, we are likely to achieve higher precision for the age composition estimates.

### **TESTING OF GENETIC REPORTING GROUPS**

Proof tests conducted by the ADF&G Gene Conservation Laboratory (GCL) demonstrated that with a fishery mixture of 100 samples, we can estimate stock composition for the four selected reporting groups (*UCI Northwest, Susitna-Matanuska, Knik-Turnagain, and Kenai Peninsula*) within 0.22 of the true values 90% of the time. These tests followed the same protocol as reported in Eskelin et al. (2013) for baseline evaluation tests; however, instead of using test mixtures with 100% of one reporting group, test mixtures were created with equal proportions of 25% from each of the four reporting groups.

### **DATA COLLECTION**

### Age, Sex, and Length Sampling

During both commercial and subsistence sampling, three scales will be removed from the preferred area of each fish and placed on an adhesive-coated card (Clutter and Whitesel 1956; Welander 1940; Appendix A). Acetate impressions will made of the scales on the card using a press under 40,000 PSI and the scale growth patterns viewed with a 40x microfiche reader to determine freshwater and marine residence times. Prior to the season, the Fisheries Biologist I will number each scale card booklet. The scale cards will be numbered consecutively, starting with 001. Each sample kit will be provided with one pre-numbered booklet. Scale cards will be pre-numbered to eliminate duplicates across sample sites. Samplers will fill out a new scale card for each sampling location and for each day. To prevent scale loss, scale cards will be stored flat between two heavy objects, with wax paper between each card. When possible, a small (approximately 7.5 cm) incision will be made from the vent forward and sex will be determined by inspection of gonads. Eggs are generally visible in ovaries which are orange in color and covered with a clear membrane whereas testes are white, flattish to triangular in cross section and composed of a solid, rubbery tissue. Sex can also be determined from external morphometric characteristics (i.e., protruding ovipositor on females or a developing kype on males). If sex cannot be easily identified, enter "U" (unknown) on the data sheet.

#### Maturity

Mature female Chinook salmon will fill and span the length of the abdominal cavity, be loosely attached in the skein with individual eggs being large and soft. Immature females will have tiny tight skeins, approximately one third to half the length of the abdominal cavity, with very small individual eggs within the skein.

Mature male Chinooks salmon will have full large soft milt sacs which also fill and span the length of the abdominal cavity. Immature males will have small tight milt sacs and also take up considerably less space in the abdominal cavity.

#### Length

MEF length will be measured to the nearest 5 mm. Chinook salmon will be sampled for ASL composition without regard to size, sex, length, or location. ASL composition data will be recorded on data sheets (Appendix B) and will follow the guidelines in (Appendix A).

#### **Tissue Sampling for MSA**

All fish sampled for ASL will also be tissue-sampled for genetic analysis. A 1<sup>1</sup>/<sub>3</sub>-cm (half-inch) piece of the axillary process will be removed from each fish and placed in a 2-ml plastic vial filled until the tissue samples are completely submerged with a Sigma<sup>1</sup> reagent grade 95% alcohol solution such that the liquid-to-tissue ratio is approximately 3:1. Sampling instructions are found in Appendix C. Each plastic vial will be sequentially numbered and vial numbers recorded on data sheets. All vials will be stored by Sport Fish and Subsistence division staff in Anchorage until the end of the season then given to the GCL for analysis.

#### **Code Wire Tag Sampling**

All Chinook salmon sampled will be examined for an adipose fin clip. Technicians will remove the head of all adipose fin clipped Chinook salmon encountered. A cinch strap will be attached to the head which will be returned to the office for storage in a freezer. All data, including the number of Chinook salmon examined and the number observed missing the adipose fin, will be recorded on a tag recovery form (Appendix D). The cinch strap number will also be recorded alongside ASL data (Appendix B) to enable cross-referencing between datasets. Data collected will be returned to the Project Leader. CWT forms and heads of all adipose fin clipped fish will be shipped at the end of the season to the ADF&G Mark, Tag, and Age Laboratory for CWT recovery, determination of stock of origin, and for archiving data.

#### **Commercial Sampling**

#### Sampling Locations

Project staff will visit as many fish processors and buyers as possible in Anchorage and Soldotna/Nikiski to collect samples from the Northern District commercial set gillnet fishery after each fishing period from late May through early June each year of the project. The fishing periods start on the first Monday on or after May 25 and continue through June 24 from 7:00 a.m. to 7:00 p.m. on Mondays unless altered by emergency order. The regular period fishing schedule is in Table 3.

<sup>&</sup>lt;sup>1</sup> Product names used in this publication are included for completeness but do not constitute product endorsement.

Sunday	Monday	Tuesday	Wednesday	Thursday	Friday	Saturday
10-May	11-May	12-May	13-May	14-May	15-May	16-May
17-May	18-May	19-May	20-May	21-May	22-May	23-May
24-May	25-May	26-May	27-May	28-May	29-May	30-May
31-May	1-Jun	2-Jun	3-Jun	4-Jun	5-Jun	6-Jun
7-Jun	8-Jun	9-Jun	10-Jun	11-Jun	12-Jun	13-Jun
14-Jun	15-Jun	16-Jun	17-Jun	18-Jun	19-Jun	20-Jun
21-Jun	22-Jun	23-Jun	24-Jun	25-Jun	26-Jun	27-Jun
28-Jun	29-Jun	30-Jun	1-Jul	2-Jul	3-Jul	4-Jul
5-Jul	6-Jul	7-Jul	8-Jul	9-Jul	10-Jul	11-Jul
12-Jul	13-Jul	14-Jul	15-Jul	16-Jul	17-Jul	18-Jul
19-Jul	20-Jul	21-Jul	22-Jul	23-Jul	24-Jul	25-Jul
26-Jul	27-Jul	28-Jul	29-Jul	30-Jul	31-Jul	1-Aug
2-Aug	3-Aug	4-Aug	5-Aug	6-Aug	7-Aug	8-Aug
9-Aug	10-Aug	11-Aug	12-Aug	13-Aug	14-Aug	15-Aug
16-Aug	17-Aug	18-Aug	19-Aug	20-Aug	21-Aug	22-Aug
23-Aug	24-Aug	25-Aug	26-Aug	27-Aug	28-Aug	29-Aug
30-Aug	31-Aug	1-Sep	2-Sep	3-Sep	4-Sep	5-Sep
6-Sep	7-Sep	8-Sep	9-Sep	10-Sep	11-Sep	12-Sep
13-Sep	14-Sep	15-Sep	16-Sep	17-Sep	18-Sep	19-Sep
20-Sep	21-Sep	22-Sep	23-Sep	24-Sep	25-Sep	26-Sep
27-Sep	28-Sep	29-Sep	30-Sep	1-Oct	2-Oct	3-Oct
4-Oct	5-Oct	6-Oct	7-Oct	8-Oct	9-Oct	10-Oct
11-Oct	12-Oct	13-Oct	14-Oct	15-Oct		

Table 3.–NCI Chinook salmon sampling schedule, 2015. Dark grey represents commercial openings and light grey represents subsistence openings.

Technicians in Anchorage will be assigned to one of three sample locations around Anchorage: 1) Ship Creek boat launch; 2) Copper River Seafoods; 3) Favco (Figure 2). Technicians in Soldotna will be assigned to one of three locations in Soldotna or Nikiski: 1) Snug Harbor Seafoods; 2) Pacific Star Seafoods; 3) Peninsula Processing (Figure 3). Because the number and location of receiving sites changes each year, the project biologist will distribute a list with contact information and maps showing locations to sample prior to the season. These will be distributed to each technician and include cell numbers (Table 4). There will be no set schedule for times to sample at each location. Schedules will depend on tides and the times of fishing periods. Deliveries may occur late at night or the day following an opener; it will be the technician's responsibility to remain in contact with the processing plant the night of and day after each opener.

A dedicated crew leader will coordinate, assist with, and oversee all sampling activities in each major geographic area. Areas are Anchorage, Soldotna/ Nikiski, and Tyonek. All staff participating in sampling will have a one day training in Anchorage or Soldotna prior to the fishing season.

#### Sampling

Technicians will collect ASL, axillary tissue, and CWT samples during each opening at receiving sites until all expected deliveries have been made. Technicians stationed at the processors will continue sampling fish as they are delivered so the location of harvest by statistical area can be determined. Samplers will identify previously sampled Chinook salmon by the lack of a left axillary appendage.

Prior to the season, the Fishery Biologist I will purchase and organize all required sampling materials into individual 10-gallon totes for each receiving site (Table 5). Throughout the season, technicians will restock and resupply these kits as needed. Prior to the 2015 season, the project biologist will arrange for the GCL to prepare pre-labeled vials.

Collection of both scale and tissue samples are expected from all Chinook salmon. However, in circumstances where the collection of scales is too time consuming and the potential exists to miss fish all together, technicians will forgo collection of scales and prioritize the collection of axillary tissue samples. Comments will be recorded when this occurs.

#### **Recording data**

Sampling crews will use a pencil to record data on waterproof datasheets (Appendix B). The header will be filled out with the date of harvest, samplers name(s), start and end times, and sampling location. The sampling location field is the name of the processor or other location at which the samples were collected. A new data sheet will be filled out for each new sampling location. The vial # will be recorded from the pre-labeled vials. Technicians will fill vials consecutively while collecting samples. Forceps will be used to collect scales from the preferred area. Scales will be mounted using the methods described in (Appendix A). Technicians will record a 'Y' to signify if scales were collected and an 'N' to signify a lack of scales. Sex of the fish will be determined from external characteristics and recorded as 'M" for male or 'F' for female; if the sex is unclear or technicians are unable to determine from external characteristics, a 'U' will be recorded to signify unknown. Chinook salmon MEF length will be measured to the nearest 5 mm using a flexible tape measure and recorded in the length column of the datasheet. The statistical area and vessel/seller name will be obtained directly from fishers or processing plants as fish arrive. The seller name is the name of the commercial fisher. Crew leaders must communicate with processor staff to prevent combining of deliveries from different statistical areas before they are sampled. If this does occur, samplers will record all applicable statistical areas.

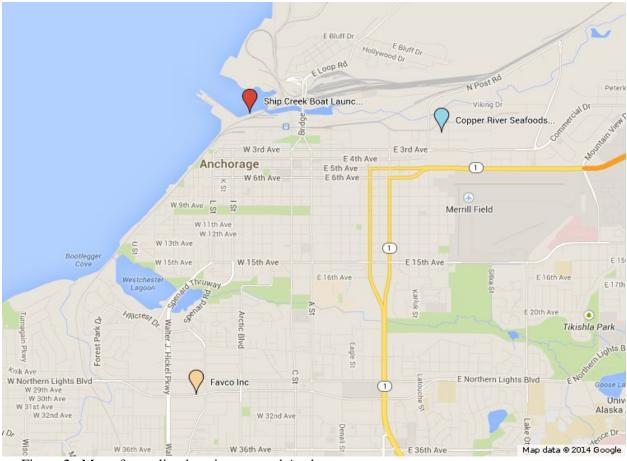


Figure 2.–Map of sampling locations around Anchorage.

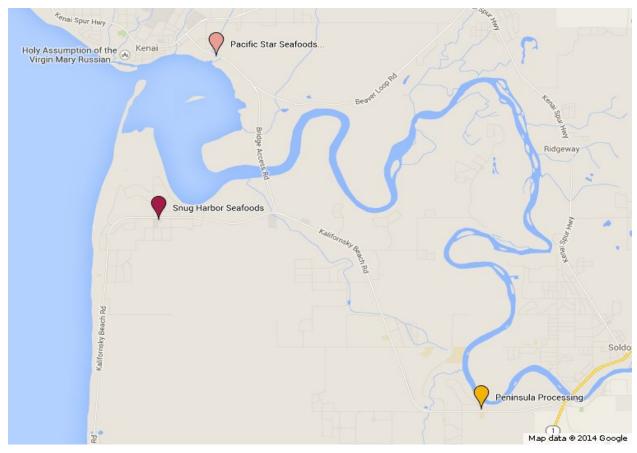


Figure 3.–Map of sampling locations around Soldotna.

#### Table 4.–NCI contact list, 2015.

Location	Division	Name	Title	Phone	comment
Soldotna	SF	Tim McKinley	FB III	260-2913	
Soldotna	SF	Tony Eskelin	FB II	260-2904	sampler
Soldotna	CF	Pat Shields	FB III	260-2941	
Soldotna	CF	Aaron Dupuis	FB II	260-2916	
Soldotna	CF	Wendy Gist	FB I	262-9368	
Soldotna	SF	Matt Sutherland	FWT		sampler
Soldotna	SF	Madeline Fox	FWT		sampler
Anchorage	SF	Sam	Info Intern		
Anchorage	SF	Adam St. Saviour	FB III	267-2170	sampler
Anchorage	SF	Jay Baumer	FB II	267-2265	sampler
Anchorage	SF	Donnie Arthur	FWT II		sampler
Anchorage	CF (GCL)	Andy Barclay	FB III	267-2290	
Anchorage	CF (GCL)	Bruce Whelan	FWT IV	267-2806	sampler
Anchorage	CF (GCL)	Charles (Hans) Thompson	FB I	267-2454	sampler
Anchorage	Subsis	Davin Holen	Program mgr	267-2807	
Anchorage	Subsis	Bronwyn Jones	SRS I	267-2178	sampler
Anchorage	Subsis	Eric	Grad Intern		sampler
Anchorage	Subsis	Dustin	Grad Intern		sampler
Anchorage	Favco	Karl or Bill		278-1525	1205 W 29th Ave
Anchorage	Favco	Karl cell			1206 W 29th Ave
Anchorage	Copper River Seafoods	Billy		522-7806	need to go in person. 1400 E 1st Ave
Soldotna	Copper River Seafoods	Clancy		335-6000	
Soldotna	Peninsula Processing	Tim or Joe		262-9678	720 Kalifornsky Beach Rd
Soldotna	Snug Harbor seafoods	Ken		283-6122	145 Kalifornski Beach Rd
Soldotna	Pacific Star Seafoods	Steve		283-7787	520 Bridge Access Rd

Item	Quantity
10 Gallon Rubbermaid tote	1
Metal clipboard	1
Instruction guide	1
Synopsis of NCI Project	5
NCI Map	1
NCI ND contacts sheet	1
Data Sheets	10
Blank notebook	1
Permanent marker	4
Pencils	4
pencil lead	1
Box pre-labled vials	3
Bullet box	1
Bag vial caps	1
EtOH	1L
Squirt bottle	1
Axillary clippers	3
Tweezers	3
Scale Card booklet (pre- numbered)	1
Roll up tape measure	2
Zak Knife	1
Small utility knife	1
paper towels	1
hand wet wipes	1
Cotton Gloves	2ea
Atlas Gloves (SM,L)	2ea

Table 5.-NCI Chinook salmon sample kit contents

#### **Subsistence Sampling**

#### Sampling Location

Subsistence fishing is open during two seasons per year. The early season, May 15 through June 15, is open for three periods per week (Tuesday, Thursday, and Friday) for 16 hours per period (4:00 a.m. to 8:00 p.m.). The late season, June 16 through October 15, is open for one period per week (Saturday) for 12 hours (6:00 a.m. to 6:00 p.m.). The regular period fishing schedule is in (Table 3). The fishery extends from a point one mile south the southern edge of the Chulitna River to the easternmost tip of Granite Point (Figure 1). Sampling will commence on May 15 and continue until the Chinook salmon run is complete, sometime around the end of June.

#### Sampling

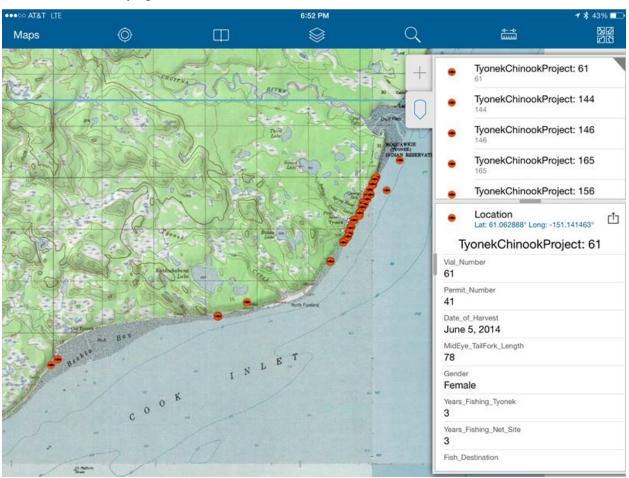
Tyonek subsistence fishery samplers will collect samples at set gillnet sites along the beach in Tyonek during the subsistence fishery. The majority of subsistence set gillnet sites are located along a one mile stretch of beach near the village center. The remaining set gillnet sites are located at Tyonek resident's family fish camps. The first camps are accessible during low to median tides via ATV along the beach. The fish camps can also be accessed with an ATV or road vehicle during high tides using the Tyonek road system. During the subsistence fishing periods, a Subsistence Resources Specialist and a Subsistence Graduate Intern will be present in the community to sample. The goal is to collect 500 samples of Chinook salmon during the season. The total community harvest can range from 600 to 1,100 annually. Early in the season 1-2 staff will be necessary to sample the harvest whereas later in the season during peak runs 3-4 staff may need to be present. All staff participating in sampling efforts were involved in the first year of the project, and will participate in a one day training in Anchorage prior to the fishing season.

#### **Recording data**

Samplers will collect a tissue sample and use an iPad data collection application to document:

- 1. Vial Number
- 2. Subsistence Permit Number
- 3. ASL data
- 4. Location based on GPS
- 5. Date of harvest
- 6. Scale card number
- 7. Number of years fishing in the Tyonek Subdistrict
- 8. Number of years at the particular net site
- 9. Ask the fisher which stream they think the fish is headed: Note if unknown
- 10. The degree of confidence they have in that location: 0=unknown, scale of 1-5
- 11. Notes on phenotypic traits which demonstrates that location
- 12. Samplers initials
- 13. Optional section to take and insert a photograph

The iPad application has a replicate feature allowing quick duplication of features, which can then be edited. The application is designed to work in rural field settings, without the need for



internet or 4G/LTE cellular data connection. Once Wi-Fi or cellular data is available, harvest data is automatically uploaded to ArcGIS online.

Figure 4.-Screen shot of iPad data collection application.

### **DATA ANALYSIS**

Technicians will return their genetic vial boxes, scale cards, and field data to their respective office daily and will be responsible for ensuring the recorded data are legible and accurate. Subsistence Division personnel will return their genetic vial boxes, scale cards, and field data to the Anchorage office upon completion of the season. The project biologist will ensure all data are returned, are legible, and are entered correctly. All data will be entered directly into a master electronic data file. Age data will be entered upon scale reading. CWT's and associated forms will be reviewed to ensure accuracy and mailed to Juneau ADF&G for entry. A final edited copy of all data files along with a data map will be sent to the Alaska Department of Fish and Game Research and Technical Services (RTS) for archiving.

## LABORATORY ANALYSIS

### Assaying Genotypes

DNA extraction and genotyping will generally follow the methods described in detail in Barclay et al. (2012). Briefly, genomic DNA will be extracted from tissue samples using a DNeasy 96 Dynamic Tissue Kit by QIAGEN (Valencia. CA). Fluidigm 192.24 Arravs (http://www.fluidigm.com) will be used to screen 48 SNP markers; this differs from the methods of Barclay et al. (2012) where 96.96 Dynamic Arrays were used. The Dynamic Arrays will be read on a Fluidigm EP1 System or BioMark System after amplification and scored using Fluidigm SNP Genotyping Analysis software. Assays that fail to amplify on the Fluidigm system will be reanalyzed on the Applied Biosystems platform. The plates will be scanned on an Applied Biosystems Prism 7900HT Sequence Detection System after amplification and scored using Applied Biosystems' Sequence Detection Software version 2.2. Genotypes produced on both platforms will be imported and archived in the GCL's Oracle database, LOKI.

### Laboratory Failure Rates and Quality Control

Overall failure rate will be calculated by dividing the number of failed single-locus genotypes by the number of assayed single-locus genotypes. An individual genotype will be considered a failure when a locus for a fish cannot be satisfactorily scored.

Quality control (QC) measures will be used to identify laboratory errors and to determine the reproducibility of genotypes. In this process, 8 of every 96 fish (1 row per 96-well plate) will be re-extracted and reanalyzed for all markers by staff not involved with the original analysis. Laboratory errors found during the QC process will be corrected, and genotypes will be corrected in the database. Inconsistencies not attributable to laboratory error will be recorded, but original genotype scores will be retained in the database.

### **BASELINE AND REPORTING GROUPS**

The current UCI Chinook salmon genetic baseline used for MSA applications is an update of the baseline reported in Barclay et al. (2012) and includes 62 additional collections and 25 new populations (Barclay et al. *In prep*; Figure 5, Table 6). The updated baseline includes the same set of SNP markers except that locus *Ots\_FGF6B* was excluded because of its association with locus *Ots\_FGF6A*.

Reporting groups take into account the likely stock contributions to the fishery by their proximities to the fishing grounds. Northern Cook Inlet fisheries are expected to contain only fish originating from Cook Inlet (therefore the Cook Inlet-only baseline). Reporting groups are defined based on one or more of the following criteria: 1) the genetic similarity among populations, 2) the expectation that proportional harvest would be greater than 5%, and 3) the applicability to answer fishery management questions. Based on these criteria, 4 reporting groups were chosen to apportion the harvest for this study (Figure 5):

- 1. *Northwest* (populations from streams in western UCI, the Yentna River, and Susitna River below the Yentna River confluence),
- 2. *Susitna/Matanuska* (populations from streams in the Susitna River above the Yentna River confluence and Matanuska River),
- 3. Knik/Turnagain (populations from spawning streams in Knik Arm and Turnagain Arm),

4. *Kenai Peninsula* (populations from spawning stream on the Kenai Peninsula from the Kenai River south to the Anchor River).

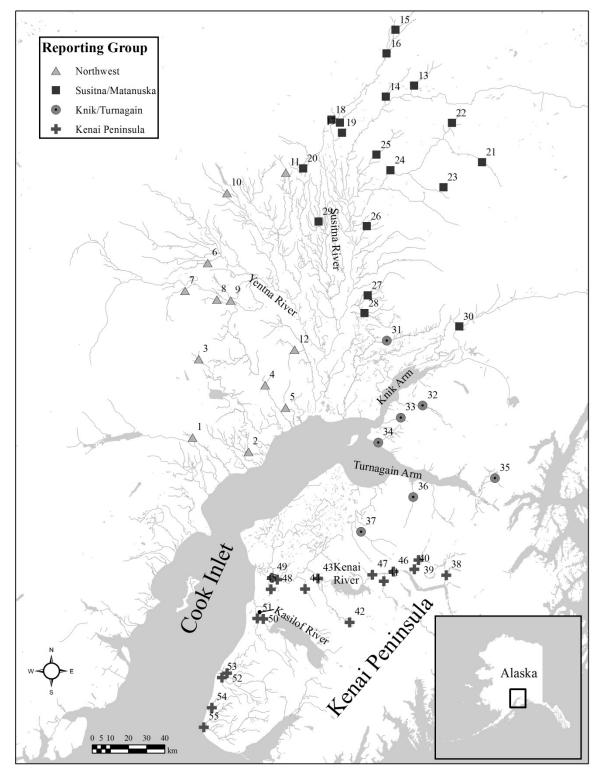


Figure 5.–Sampling locations for Chinook salmon originating for Upper Cook Inlet, Alaska, 1995–2013 and reporting groups that will be used for mixed stock analysis of the Tyonek subsistence fishery and Northern District commercial fishery harvests. Numbers correspond to map numbers on Table 6.

Map			Added after		
No	Reporting Group	Location	baseline <sup>a</sup>	Collection year(s)	N
1	Northwest	Straight Creek		2010	95
2		Chuitna River		2008, 2009	134
3		Coal Creek		2009, 2010, 2011	118
4		Theodore River	Х	2010, 2011, 2012	190
5		Lewis River	Х	2011, 2012	87
6		Red Creek	Х	2012, 2013	111
7		Hayes River	Х	2012, 2013	50
8		Canyon Creek	Х	2012, 2013	91
9		Talachulitna River		1995, 2008, 2010	178
10		Sunflower Creek		2009, 2011	123
11		Peters Creek	Х	2009, 2010, 2011, 2012	107
12		Sucker Creek	Х	2011, 2012	143
13	Susitna/Matanuska	Portage Creek	Х	2009, 2010, 2011, 2013	162
14		Indian River	Х	2013	79
15		Middle Fork Chulitna River	Х	2009, 2010	169
16		East Fork Chulitna River	Х	2009, 2010, 2011, 2013	77
17		Byers Creek	Х	2013	55
18		Spink Creek	Х	2013	56
19		Troublesome Creek	Х	2013	71
20		Bunco Creek	Х	2013	98
21		no name creek	Х	2013	69
22		Prairie Creek		1995, 2008	161
23		East Fork Iron Creek	Х	2013	57
24		Disappointment Creek	Х	2013	64
25		Chunilna Creek		2009, 2012	123
26		Montana Creek		2008, 2009, 2010	213
27		Little Willow Creek	Х	2013	54
28		Willow Creek		2005, 2009	170
29		Deshka River		1995, 2005, 2012	303
30		Moose Creek		1995, 2008, 2009, 2012	149
31	Knik/Turnagain	Little Susitna River		2009, 2010	125
32		Eagle River	Х	2009, 2011, 2012	71
33		Ship Creek		2009	261
34		Campbell Creek	Х	2010, 2011, 2012	110
35		Carmen River	Х	2011, 2012	50
36		Resurrection Creek	Х	2010, 2011, 2012	98

Table 6.--Populations of Chinook salmon in the Upper Cook Inlet genetic baseline, including the sampling location, collection years, the number of individuals sampled from each population (N), and the reporting groups for mixed stock analysis of the Tyonek subsistence fishery and Northern District commercial fishery harvests.

37		Chickaloon River		2008, 2010, 2011	128
38	Kenai Peninsula	Grant Creek	Х	2011, 2012	55
39		Quartz Creek		2006, 2007, 2008, 2009, 2010, 2011	131
40		Crescent Creek		2006	164
41		Russian River		2005, 2006, 2007, 2008	214
42		Benjamin Creek		2005, 2006	204
43		Killey River		2005, 2006	255
44		Funny River		2005, 2006	219
45		Slikok Creek		2004, 2005, 2008	136
46		Juneau Creek		2005, 2006, 2007	140
47		Upper Kenai River mainstem		2009	191
48		Middle Kenai River mainstem		2003, 2004, 2006	299
49		Lower Kenai River mainstem	Х	2010, 2011	118
50		Kasilof River mainstem		2005	321
51		Crooked Creek		2005, 2011	306
52		Ninilchik River Weir		2006, 2010	209
53		Deep Creek		2009, 2010	196
54		Stariski Creek	Х	2011	104
55		Anchor River Weir		2006, 2010	249

*Note:* Map numbers correspond to sampling sites on Figure 1.

<sup>a</sup> "X" indicates populations that have been added since the Barclay et al. (2012) baseline.

#### MIXED STOCK ANALYSIS

The stock composition of the Tyonek subsistence and Northern District commercial fisheries harvests for each stratum will be estimated using the software package BAYES (Pella and Masuda 2001). BAYES employs a Bayesian algorithm to estimate the most probable contribution of the baseline populations to explain the combination of genotypes in the mixture sample. The final analysis will consist of the results from 5 separate Monte Carlo Markov chains where each chain will begin with different initial values. A random number generator will be used to create the initial values which will sum to 1.0 over all reporting groups. The Dirichlet prior distribution for the composition parameters in BAYES will be based upon the best available information for each mixture analysis. We believe the best available information for the prior to be the results of MSA of similar mixtures. Because no MSA results are available for similar mixtures, the Dirichlet prior distribution for the first temporal stratum in each fishery will be defined to be equal (i.e., a "flat" prior) with the prior for a reporting group divided equally among populations within that reporting group for population prior parameters. For the analysis of subsequent temporal strata, the priors will be defined as the posterior means of the previous temporal stratum. The sum of the Dirichlet prior parameters will equal 1, thus minimizing the overall influence of the prior distribution. The chains will be run until convergence is reached (shrink factor < 1.2) for the 5 chains (Pella and Masuda 2001). The first half of each chain will be discarded in order to remove the influence of the initial values; the rest will be used to estimate the posterior distribution of stock composition proportions. The point estimates of stock composition and the variance of these estimates will be calculated from the mean and standard deviation of the posterior distributions.

### HARVEST OF CHINOOK SALMON BY REPORTING GROUP

The number of Chinook salmon from reporting group  $g(\hat{H}^s)$  harvested in the UCI Northern District commercial fishery between the first opening as early as late May and the last opening on or before June 24 will be estimated as:

$$\hat{H}^{g} = \sum_{i=1}^{T} \sum_{j=1}^{S} H_{i,j} \hat{p}_{i,j}^{g}$$
(1)

where

- $\hat{p}_{i,j}^{g}$  = estimated proportion of NCI harvest in time stratum *i* and geographic stratum *j* comprising Chinook salmon from reporting group *g* (*Northwest, Susitna/Matanuska, Knik/Turnagain,* or *Kenai Peninsula*). Obtained based on Bayesian mixed stock analysis as described in the previous section.
- $H_{i,j}$  = NCI Chinook salmon harvest in time stratum *i* and area stratum *j* obtained from fish ticket data.

$$T =$$
 number of time strata (25 May - 15 June, and 16-30 June)

S = number of geographic strata (Trading Bay/ Pt. McKinzie, Susitna Flats, and Fire Island/ Eastern Sub-district)

 $var(\hat{H}^{g})$  will be estimated as:

$$\operatorname{var}(\hat{H}^{g}) = \sum_{i} \sum_{j} (H_{i,j})^{2} \operatorname{var}(\hat{p}_{i,j}^{g})$$
(2)

where  $\operatorname{var}(\hat{p}_{i,j}^{g})$  will be available from the Bayesian mixed stock analysis (Pella and Masuda, 2001).

Harvest of Chinook salmon by reporting group  $g(\hat{H}^s)$  from the Tyonek subsistence fishery will be estimated using equations (1) and (2) with the appropriate substitutions for the Tyonek fishery.

### AGE, SEX, AND MATURITY COMPOSITION OF CHINOOK SALMON

For clarity, the following description and formulae were developed in terms of estimating the age composition, however estimating the sex composition is treated identically. The age proportions of Chinook salmon harvested in the Tyonek subsistence and Northern District commercial fishery by sampling stratum will be estimated as:

$$\hat{p}_{i,j}^{z} = \frac{n_{i,j}^{z}}{n_{i,j}}$$
(3)

where  $\hat{p}_{i,j}^{z}$  is the estimated proportion of salmon of age category z from sampling stratum (i, j),  $n_{i,j}^{z}$  equals the number of fish sampled from sampling stratum (i, j) that were classified as age category z, and  $n_{i,j}$  equals the number of Chinook salmon sampled for age determination from sampling stratum (i, j).

The variance of  $\hat{p}_{i,j}^{z}$  will be estimated by:

$$\operatorname{var}[\hat{p}_{i,j}^{z}] = \left(1 - \frac{n_{i,j}}{H_{i,j}}\right) \frac{\hat{p}_{i,j}^{z} \left(1 - \hat{p}_{i,j}^{z}\right)}{n_{i,j} - 1}$$
(4)

where  $H_{i,j}$  is the number of Chinook salmon harvested in a sampling stratum (i, j).

The estimates of harvest by age categories in each sampling stratum will be calculated by:

$$\hat{H}_{i,j}^{z} = H_{i,j}\hat{p}_{i,j}^{z}$$
(5)

with its variance estimated as:

$$\operatorname{var}\left[\hat{H}_{i,j}^{z}\right] = H_{i,j}^{2} \operatorname{var}\left[\hat{p}_{i,j}^{z}\right]$$
(6)

The total harvest by age category and its variance will then be estimated by summation:

$$\hat{H}^{z} = \sum_{i=1}^{T} \sum_{j=1}^{S} \hat{H}_{i,j}^{z} \qquad \text{var} \Big[ \hat{H}^{z} \Big] = \sum_{i=1}^{T} \sum_{j=1}^{S} \text{var} \Big[ \hat{H}_{i,j}^{z} \Big]$$
(7)

where: T=3, S=2 are the number of time and geographic strata respectively.

Finally, the total proportion of the Tyonek subsistence or Northern District commercial harvest by age category and its variance will be estimated by:

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$$\hat{p}^{z} = \frac{\hat{H}^{z}}{H} \qquad \qquad \operatorname{var}[\hat{p}^{z}] = \frac{\operatorname{var}[\hat{H}^{z}]}{H^{2}} \tag{8}$$

#### **CWT RECOVERIES**

With the low numbers of CWT recoveries expected, no direct estimates of CWT recoveries by stock will be made, but the data will be archived with Mark, Tag, and Age Laboratory in Juneau.

## **BUDGET SUMMARY**

FY16		
Line Item	Catagory	Budget
Line item	Category	(x\$1,000)
100	Personnel	119.0
200	Travel	6.6
300	Contractual	5.6
400	Commodities	1.0
500	Equipment	0.0
Total		132.2

## SCHEDULE AND DELIVERABLES

Date	Activity
February – Mid May 2015	Hiring and preseason training (St. Saviour) DNA extracted by GCL from samples collected in 2014
Mid May – Late June 2015	NCI Chinook salmon harvest sampling (10 FWT)
September 2015	Data edited, tissue collection transferred to GCL, and CWT forms and heads mailed t Juneau Mark, Tag, and Age lab (St. Saviour and Jalbert)
October 2015	Scales aged, ASL composition estimates completed (Jalbert)
July 2016	Tissue collection transferred to GCL and DNA extracted from samples collected in 2016, DNA extractions from 2014–2016 genotyped by GCL
August 2016	MSA completed and results disseminated (Barclay)
September 2016	Harvest estimates completed by temporal, geographic strata and reporting group. (Antonovich)
October – November 2016	Draft peer-reviewed journal article; submit final report to AKSSF (St. Saviour, Barclay, Antonovich)

## RESPONSIBILITIES

### **PRINCIPAL INVESTIGATOR**

Adam St. Saviour, Project Leader, Fishery Biologist III:

The project leader is responsible for analysis, reporting, and budget oversight. This position will serve as the project biologist and will be responsible for hiring and training personnel and supervision of data collection. The project biologist will plan, coordinate, and participate in field sampling. This position will also ensure all data is in proper format and archived with RTS at the completion of the field season and will be primary author on any reporting.

### **CO-PRINCIPAL INVESTIGATORS**

Andrew Barclay, Fishery Biologist III:

This position is the Gene Conservation Lab representative. This position is responsible for the analysis of tissue samples for MSA, and providing estimates to the project biologist and biometrician. This position will be co-author on FDS reports and memos.

Davin Holen, Subsistence Program Manager, Southern Alaska:

This position is responsible for the designing the methodology for data collection in the subsistence fishery, and assisting with analysis, reporting, and budget oversight. This position provides oversight duties for Division of Subsistence research staff.

### **CONSULTING BIOMETRICIAN**

Anton Antonovich, Biometrician III:

This position will provide guidance on sampling design and data analysis, prepare estimates of harvest of Chinook salmon by reporting group and assist with preparation of the operational plan and any reports.

#### **PROJECT ASSISTANTS**

Chase Jalbert, Fishery Biologist I:

This position will directly assist the Principal Investigator with project implementation. The assistant's responsibilities will include assisting with writing the operational plan, purchasing and preparing field equipment, training new technicians, coordinating sampling efforts, sampling Chinook salmon for ASL and tissue, reading scales, entering data, organizing and distributing tissue samples to GCL.

Bronwyn Jones, Subsistence Resource Specialist I:

This position will directly assist the Principal Investigator with project implementation. The assistant's responsibilities will include coordinating the Tyonek sampling schedule, training subsistence personnel and field supervision of Tyonek data collection.

### SAMPLING CREWS

Fish and Wildlife Technicians III (~3 part-time):

Conduct sampling of Chinook salmon for ASL and tissue. Ensure sampling procedures are in accordance with the operational plan. Conduct quality control on collected field data.

Fish and Wildlife Technicians II (~5 part-time):

Responsibilities of these positions include: Operating State of Alaska vehicles, adhering to sampling schedule; sampling harvested Chinook salmon for ASL and tissue; recording data accurately; entering data into a computerized database in a timely manner.

Graduate Interns, Division of Subsistence (~2 full-time):

Responsibilities of these positions include: Operating State of Alaska vehicles, adhering to sampling schedule; sampling harvested Chinook salmon for ASL and tissue, and documenting harvest locations and local knowledge of salmon. These positions also assist in data collection in the commercial fishery.

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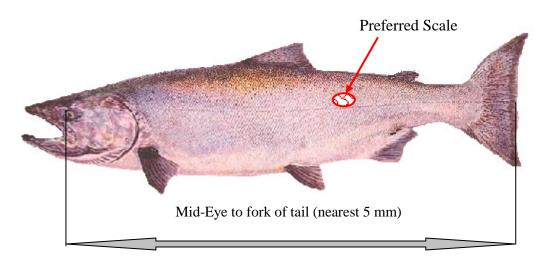
## APPENDIX A.-ADULT SALMON ASL SAMPLING

### SAMPLING PROCEDURES

Place the salmon flat on its right side (the head should be toward the left).

#### Measure the length (in mm)

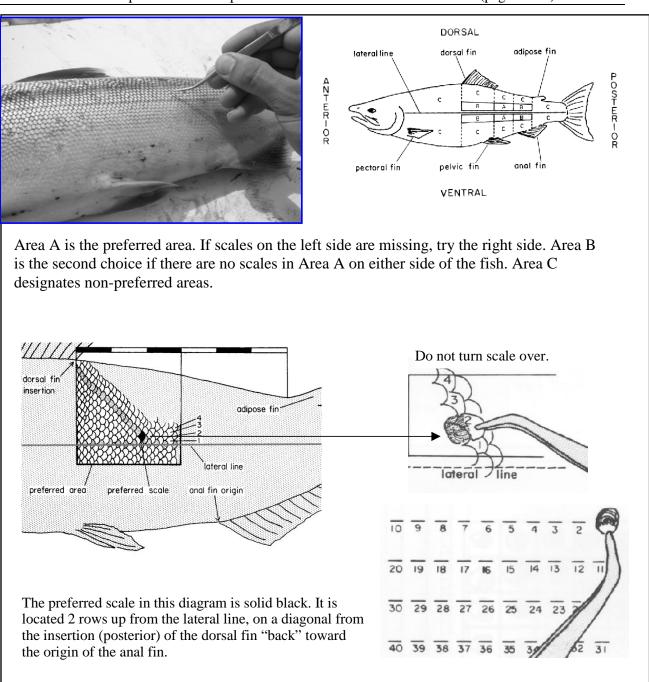
Adult salmon length is measured from mid-eye to tail fork because the shape of the salmon's snout changes as it approaches sexual maturity. With your left hand, place the tape measure so the end starts in the middle of the eye and runs back towards the tail. Flatten and spread the tail and pull the tape tight with your right hand. Read and record the mid-eye to tail fork length to the nearest 5 mm.



**Determine the sex** of the fish using external characteristics (protruding ovipositor on females or a developing kype on male).

#### Remove the preferred scale and place on scale card

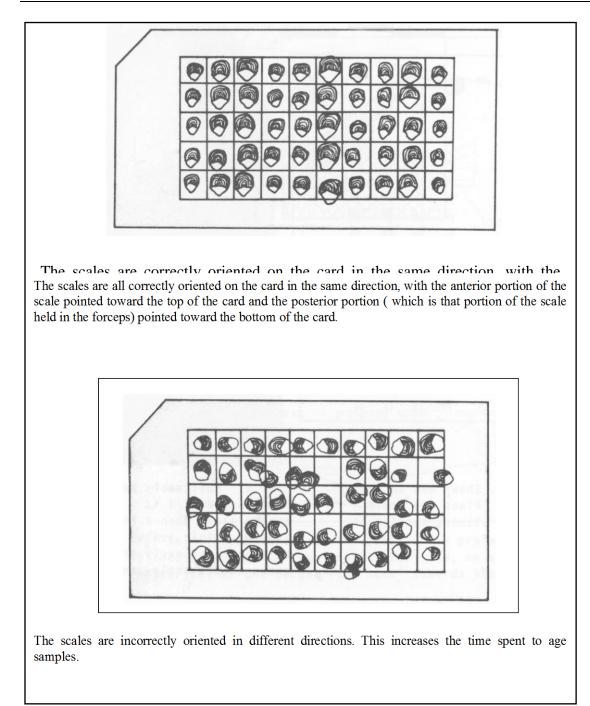
When sampling Chinook salmon, four scales are taken from the preferred area, cleaned of all grit and slime by rubbing between the thumb and forefinger and properly placed on a labeled scale (gum) card (Figures 2 and 3). The scales for fish #1 will be mounted over the numeral "1,11,21,31", continuing to mount the scales from fish #2 over the numerals "2,12,22,32" and so on. Samplers should be careful to make sure that the scale is not flipped over before it is placed on the scale card.



A2.–Removal and placement of the preferred salmon scale onto the scale card (page 2 of 3)

-continued-

A3.–Scale orientation on scale card (page 3 of 3)



# **APPENDIX B.- NCI CHINOOK SALMON SAMPLING FORM**

			NCI C	hinook Salmor	Sampling F	orm
Date:          Sampler(s):            Start Time:          End Time:						
				End Time:		
				processor):		
	vpe: <u>Setne</u>					
				Fin Clip: Axillary		
Vial #				Catch Location (Statistical Area)		Comments
-						
			1			
Carles	 				 	
scales:	iviount 4 scal	ies troi	m preferr	ed area on a scale c	aiu;	Length: mid eye to fork-of-tail

## APPENDIX C.-INSTRUCTIONS FOR TISSUE SAMPLING

#### Non-lethal Sampling Finfish Tissue for DNA Analysis ADF&G Gene Conservation Lab, Anchorage

#### I. General Information

We use axillary tissue samples from individual fish to determine the genetic characteristics and profile of a particular run or stock of fish. The most important thing to remember in collecting samples is that **only quality tissue samples give quality results**. If sampling from carcasses: tissues need to be as "fresh" and as cold as possible and recently moribund, do not sample from fungal fins.

## Preservative used: Isopropanol/Methanol/Ethanol (EtOH) preserves tissues for later DNA extraction. Avoid extended contact with skin.

#### II. Sampling Method



located above pelvic fin. Each clip should maintain a ratio 3 EtOH/1 axillary "spine" in vials for best results. Using clipper; cut ½ - 1" max.



#### III. Supplies included with sampling kit:

- 1. Dog toe nail clipper for cutting a portion of the axillary process.
- 2. Cryovials 2.0ml pre-labeled plastic vial or tube.
- 3. Caps cap for each vial.
- 4. Cryovial rack- white plastic rack with holes for holding cryovials while sampling.
- 5. Ethanol (EtOH) in Nalgene bottle(s).
- 6. Squirt bottle to fill and/or "top off" each cryovial with EtOH
- 7. Laminated "return address" labels
- 8. Sampling instructions

#### IV. Shipping: HAZMAT paperwork is required for return shipment of these samples.

Return to ADF&G Anchorage lab:	ADF&G – Genetics 333 Raspberry Road	Lab staff: 907-267-2247 Judy Berger: 907-267-2175	
	Anchorage, Alaska 99518	Freight code:	

- Wipe excess water and/or slime off the axillary process "spine" prior to sampling to avoid getting either water or fish slime into the 2.0ml vial (see diagram).
- Prior to sampling, fill the tubes half way with EtOH. Fill only the tubes that you will use for each sampling period. The squirt bottle is for day use only since it will leak overnight when unattended.
- Clip off the axillary "spine" using dog nail clippers or scissors to get roughly a <sup>1</sup>/<sub>2</sub> - 1" inch maximum piece and/or about the size of a small fingernail.
- Place axillary process into EtOH. The ethanol/tissue ratio should be **slightly less than 3:1** to thoroughly soak the tissue in the buffer.
- Top up tubes with EtOH and screw cap on securely. Invert tube twice to mix EtOH and tissue. Periodically, wipe or rinse the clippers with water so not to cross contaminate samples.
- Data to record: Record each vial number to paired data information (i.e. location, lat./long., sample date(s), etc.). Electronic version preferred.
- Discard remaining ethanol from the 500ml bottles before shipping. Tissue samples must remain in 2ml EtOH, these small quantities require HAZMAT paperwork. Please follow packing instructions for HAZMAT items. Store vials containing tissues at room temperature, but away from heat. In the field: keep samples out of direct sun, rain and store capped vials in a dry, cool location. Freezing not required.

## APPENDIX D.-CODED WIRE TAG SAMPLING FORM



# Alaska Department of Fish and Game

	Comme	cial Fisher	Sampling Fo ies stward & AYK F		Pro 1 31	Page Info tor inta angle Number only! See Instructions	OF OF
	SAMPLE NUMBER:	1				DATE FIRST C	
Z	HARVEST TYPE: 11-traditional 21-pnp-fis	ı	SURVEY SITE:				
∆TK	12-terminal-area 22-pnp-ca 13-exper-area 41-test-ru 18-confiscated 42-test-sp	i-strength	SAMPLE TYPE:	random	select	DATE SOLD (L	- 1
$\geq$	10-00111302100 42-1031-3	strai	SAMPLE TIME:		end		- 1
R	PROCESSOR:	MATION	Lower Cook Inlet		-		Г) <u>ҮК</u> 31-
	BUYING STATION:           ADF&G#:           VESSEL OR		232- 241-	(Invalid Subdistricts) 244-20, -30, -40 245-	252- 253-	257- 33 258- ot	34- THER DISTRICTS
Ζ	OWNER'S NAME:		246- 247- FISHED:	254- 255-	259 262		
	GEAR 01-purse seine 0 TYPE: 03-drift gillnet 0 08 - fish wheel	WATER TYPE: ANADROMOUS STREAM# (FRESHWATER- ONLY)		freshwater		<sup>.</sup>	
IN	THIS BOX IS TO BE COMPL FOR RANDOM SAME	TED ONLY		SPE	CIES LENG	TH NOTES	ed) (Chin.
Z	TOTAL # FIBH CHECKED : SPECIES FOR AD- (CODE) AD-CLIPS SE	WERE JPS ALL N CHECKED?					
	(410)CHIN	y n					
Ż	(420)SOCK	y n					
	(430)COHO (440)PINK	y n y n		+	+		
	(450)CHUM	y n					
	COMMENTS:	y n					
54 P					T:FOR	M8/2011/18/0/COMCWA20	011V8D 03/24/11 01:35