Dr. Robert Behnke Department of Fishery & Wildlife Biology Colorado State University Fort Collins.Colorado 80523

Dear Bob:

I apologize for being so slow in answering your letter. I've been on the road lately and heavily involved in the Yukon River negotiations between the U.S. and Canada.

We've looked at a number of char populations across the North Slope of Alaska and Canada the past few years for a genetic stock identification study. I've enclosed a draft copy of our report. Unfortunately, we just haven't had the time or resources to do any serious comparisons with any Dolly Varden populations. We are currently doing electrophoretic work on approximately 8,000 samples from the North Slope, Yukon River, and Bristol Bay. As a result, we've been rather overwhelmed. We did look at some Dolly Varden samples from a couple of interior rivers and from Karluk Lake a few years ago and found no fixed differences between these samples and the North Slope char. There were, however, some major frequency differences.

We hope to get a short breather this winter and plan to write up a couple of publications on the char work - one on just the electrophoretic results with comparisons with Dolly Varden and other char populations, and a second on the genetic stock identification results. When we get a manuscript together, I will send you a copy for review.

On another note, we are having difficulty finding someone to fill a GS-9 Geneticist position. We recent advertized the position and had no takers. If you know of anyone who might be interested, please have them contact me. We are re-advertizing the position and I've enclosed the announcement.

I hope we can get back to you this winter with some more definitive comparisons between the Arctic char and Dolly Varden. Have a good holiday season.

Sincerely,

Richard L. Wilmot

POPULATION GENETIC STRUCTURE OF ARCTIC CHAR (Salvelinus alpinus) FROM RIVERS OF THE NORTH SLOPE OF ALASKA AND CANADA

> Rebecca J. Everett Richard L. Wilmot

12/7/88 Dimit

Dean W. Cramer

· 1988

Part of a joint study, entitled: Arctic Fish Habitats and Sensitivities

by

Alaska Fish and Wildlife Research Center

and

Fisheries Management Services

U.S. Fish and Wildlife Service 1011 East Tudor Road Anchorage, Alaska 99503

Report of research funded by:

U.S. Minerals Management Service

#### through

National Oceanic and Atmospheric Administration

1	Page
List of Tables	v
List of Figures	vi
Abstract	vii
Technical Summary	iii
Introduction	1
Background	3
Objectives	5
Methods	7
Baseline Collections	7
Offshore Collections	7
Electrophoresis	10
Statistical Methods	12
Amount of Genetic Variation	12
Genotypic Distribution	12
Genetic Heterogeneity	13
Genetic Similarity	14
Gene Diversity Analysis	15
Genetic Stock Identification	16
Simulations	16
Endicott Samples	17
Results	18
1987 Collections	18
Amount of Genetic Variation	19

	Page
Genotypic Distributions	21
Genetic Heterogeneity	21
Genetic Similarity	22
Gene Diversity Analysis	25
Genetic Stock Identification	25
Artificial Mixed Stock Simulations	25
Artificial Mixture	29
Incremental Simulations	31
Twenty-Percent Incremental	31
Mixed Fishery SamplesEndicott	39
Discussion	43
Collections	43
Amount of Genetic Variation	44
Combining Baseline Data	45
Genetic Differences Among Populations Within Drainages	48
Genetic Divergence Among Populations	50
Genetic Stock Identification: Simulations	52
Evaluating Genetic Stock Divergence	55
Genetic Stock Identification: Endicott Samples	56
Implications for Management	60
Literature Cited	63

		Page
Appendix A:	Gene frequencies of variable loci in 12 populations	
	of Arctic char collected in 1987 from the North	
	Slope of Alaska and Canada	68
Appendix B:	Gene frequencies of variable loci in 16 populations	
	of Arctic char collected in 1986 and 1987 from the	
	North Slope of Alaska and Canada	69
Appendix C:	Mean estimate (with one bootstrap standard	
	deviation) and actual proportion of an artificial	
	mixed stock analyzed with maximum likelihood method	
	of genetic stock identification	70
Appendix D:	Percentage allocations to each of 14 char stocks	
	when an artificial mixtures of each stock is	
	compared to baseline data using the maximum	
	likelihood method of genetic stock identification	71
Appendix E:	Mean estimates (with one bootstrap standard	
	deviation) of the composition of the June sample	
	of Arctic char (N = $208$ ) taken from near the	
	Endicott causeway	72
Appendix F:	Mean estimates (with one bootstrap standard	
	deviation) of the composition of the July sample	
	of Arctic char (N = 126) taken from near the	
	Endicott causeway	73

iii

Appendix G: Mean estimates (with one bootstrap standard deviation) of the composition of the August sample of Arctic char (N = 126) taken from near the Endicott causeway - - - - - - - - - - 74

iv

Page

# LIST OF TABLES

v

Table 1.	Populations of Arctic char collected from rivers of	
	the North Slope of Alaska and Canada with site location,	
	number of samples collected from each site, and date	
	of collection	8
Table 2.	Enzymes, Enzyme Commission numbers, and loci examined	
	in samples of Arctic char collected from northern	
	Alaska in 1987	11
Table 3.	Expected average percent of fish heterozygous per locus,	
	and percent of loci examined that were polymorphic in	
	16 populations of Arctic char sampled from rivers of	
	the North Slope of Alaska in 1986 and 1987	20
Table 4.	Log likelihood ratio heterogenity tests among	
	populations sampled at different sites and/or	
	different years	23
Table 5.	Matrix of genetic heterogeneity, tested pairwise	
	among Arctic char populations of the North Slope,	
	Alaska and Canada	24
Table 6.	Matrix of Nei's (1978) gene identity values pairwise	
	among 16 populations of Arctic char sampled from the	
	North Slope of Alaska rivers in 1986 and 1987	26
Table 7.	Gene diversity analysis (Nei 1973; Chakraborty 1980)	
	among populations of Arctic char from rivers of the	
	North Slope of Alaska and Canada	28

Page

# LIST OF FIGURES

		Page
Figure 1.	Sampling sites for Arctic char genetics research	. 9
Figure 2.	Relationships based on Nei's (1978) index of genetic	
	identity of 16 populations of Arctic char from rivers	
	of the North Slope of Alaska and Canada	- 27
Figure 3.	Maximum likelihood method of genetic stock	
	identification on an artificial mixed stock of North	
	Slope Arctic char	- 30
Figure 4.	Incremental artificial mixed stock simulations for	
	each of 14 populations of North Slope Arctic char	- 32
Figure 5.	Incremental artificial mixed stock simulations for	
	each of 14 populations of North Slope Arctic char	- 35
Figure 6.	Estimated composition (±1.28 standard deviations)	
	of a mixed fishery sample from Prudhoe Bay, Alaska	
	collected in June 1987	- 40
Figure 7.	Estimated composition (±1.28 standard deviations)	
	of a mixed fishery sample from Prudhoe Bay, Alaska	
	collected in July 1987	- 41
Figure 8.	Estimated composition (±1.28 standard deviations)	
	of a mixed fishery sample from Prudhoe Bay, Alaska	
	collected in August 1987	- 42

DRAFT

#### INTRODUCTION

1

Potential problems with oil exploration and development on the North Slope of Alaska include the effects of offshore causeways, other structures, and development activities on anadromous species such as Arctic char (<u>Salvelinus alpinus</u>) and Arctic cisco (<u>Coregonus</u> <u>autumnalis</u>). In offshore areas, these species are in mixed aggregations, making it difficult to determine specifically which stocks could be at risk during what time period. In the tributaries, development activities such as river crossings or water removal from pools vital to overwintering could also affect migration, spawning, or rearing areas.

Arctic char are of special interest because of their relatively low abundance, limited range in a narrow band of coastal waters, and their importance in subsistence fisheries. Studies (e.g., Furniss 1975; Craig and McCart 1975) characterizing the marine and freshwater phases of Arctic char life history emphasize the importance of the coastal area of the Beaufort Sea. Populations of Arctic char migrate from freshwater, springfed spawning and overwintering areas to nearshore marine feeding grounds. Migration studies using mark and recapture techniques indicate that movements are generally limited to an area adjacent to the river of origin (Furniss 1975). However, examples of extended migration and overwintering in drainages other than those used for spawning have been documented (Craig and McCart 1976; Glova and McCart 1974).

In order to determine which stocks could be affected by

development, and possibly to predict how they would be affected, we need to understand the stock structure of both the inland and offshore stages of the Arctic char life history. By studying the amount and pattern of genetic variation in the populations while they are associated with their natal drainages, we can make inferences about

the evolutionary history of northern Arctic char, and predict their ability to respond to changing environmental conditions. Using samples from spawning populations of the tributaries, we can genetically characterize these individual stocks, determine their relationships to each other, and form a baseline.

2

With baseline information, we can estimate the composition of mixed stocks using genetic stock identification methods to compare the "genetic types" of the baseline to the mixed stock samples. Electrophoretic detection of protein variation makes it possible to discriminate among stocks using quantifiable characters having a genetic basis.

Electrophoretically distinguishable characters have generally proven to be stable characteristics of fish stocks that have been studied (see Utter et al. 1980; Grant et al. 1980; Milner et al. 1980; Campton and Utter 1987; and Robin Waples, National Marine Fisheries Service, personal communication). Exceptions have been observed in Alaska sockeye (Wilmot, U.S. Fish and Wildlife Service, unpublished data) and in chinook of the Columbia River, Washington (Milner et al. 1980). In the latter case, shifts apparently related to straying from transplantation and migration, though selection is a possible explanation.

DRAFT

If the species of concern has a suitable stock structure, biochemical genetics methods can be used to estimate the percent composition of various stocks represented in mixed aggregations sampled from offshore areas. This type of information can provide site-specific information on stocks at risk from habitat alteration, and biological data at migratory stages of the life cycle of these natural populations.

#### Background

The objectives of the first year of this study were to: 1) characterize the amount and pattern of genetic variation in populations of anadromous Arctic char from major drainages of the North Slope of Alaska, 2) determine whether the population structure of North Slope char is such that genetic stock identification of mixed populations collected from offshore waters would be possible, and 3) describe how a sampling program would be designed to use genetic stock identification to determine which stocks would be affected by specific development projects.

In 1986, samples of juvenile Arctic char from 15 populations were collected from 10 tributaries to the Beaufort Sea. We used horizontal starch-gel electrophoresis to identify protein products of 42 loci coding for 20 enzymes in three tissues. We measured the amount of variation, the pattern of variation (genotypic distribution) within population samples, the similarity between populations, their heterogeneity, and the degree of gene diversity among groups.

3

We found that northern Alaska Arctic char have more genetic variation than might be expected given the relatively narrow range of waters they inhabit and the harsh environmental conditions. With an average heterozygosity per locus of 5.1%, they are typical of fish species in general; at the upper end of the range observed in other salmonid fishes; and higher than most other Arctic char populations that have been studied.

4

The genetic identities (Nei 1972; 1978) among riverine North Slope Arctic char populations studied are high (>0.987), indicating fairly recent common ancestry. High similarity values do not imply lack of significant differences between populations. Heterogeneity tests between populations indicated the distinctness of the populations and the complexity of the relationships between them. Almost all North Slope Arctic char populations studied are significantly genetically distinct from each other. The implication of this information is that fish from different drainages are reproductively isolated, and are most likely true to their spawning streams.

Since anadromous char do mix to some unknown degree in feeding areas, the differences that have been established between stocks are likely maintained by homing behavior. Populations of each drainage are probably discrete, locally adapted units. It is not clear at this time how non-migratory forms are related to anadromous stocks. Various authors maintain differences within drainages.

It is unlikely that loss of any one stock would be mitigated by substitution of another. While the actual loci we have studied may be selectively neutral, underlying variation that is marked by these loci

may be highly selected for in different environments, corresponding generally to different drainages. As such, Arctic char stocks of the North Slope should be managed as individual, unique gene pools.

5

North Slope Arctic char do not have the magnitude of difference between groups exhibited by the non-migratory char of northern Europe. They do, however, compare with the population structure of anadromous Pacific salmon. To do genetic stock identification there must be sufficient detectable genetic variation between populations of different major drainages, combined with a low within-group variability.

Our data (Everett and Wilmot 1987) indicate that North Slope char have a relatively large amount of genetic variation; there are significant differences among populations; and the observed variation is partitioned such that there is as much difference between char from different drainages as there is among populations of sockeye and chum salmon where genetic stock identification has been used successfully. As such, we could anticipate successful application of this technique to the identification of char at specific offshore sites, and designed a project to collect additional samples for baseline data, plus offshore samples from Beaufort Sea mixed aggregations near Endicott Causeway, Prudhoe Bay, and Camden Bay.

#### **Objectives**

The goals of the 1987 Arctic Fish Habitats and Sensitivities Study are to understand the genetics of the anadromous North Slope Arctic

11/29/88

char populations, and to determine which of the populations using certain offshore areas of the Beaufort Sea would be affected by development projects in that area.

6

The objectives of the genetic stock identification study are to: 1) analyze additional populations of Arctic char that are major contributors to the offshore mixed stock of the Beaufort Sea, 2) compare the Arctic char of the drainages of the Chukchi Sea to those of the Beaufort Sea, and 3) collect samples of Arctic char from the mixed stock in the Beaufort Sea and estimate the percent composition of fish from the baseline populations we have studied that contribute to mixed stock collections.

To accomplish these objectives, we collected samples of Arctic char from several additional river sites for baseline information, and acquired samples from mixed stocks at the mouth of the Sagavanirktok Drainage at the Endicott Causeway and from the Camden Bay area. We did electrophoretic analysis of protein variation for all these fish, and did statistical analyses to determine the relationships of the collections to each other (over space, time, and resident versus non-resident populations). We then used the baseline data to do computer simulations to determine the accuracy and precision of our database, and then did actual stock-composition analyses of the samples from the Endicott area.

11/29/88

DRAFT

#### METHODS

7

#### Baseline Collections

U.S. Fish and Wildlife Service crews used electroshockers and minnow traps to sample for Arctic char at 22 sites on 16 rivers. Char were found at 12 sites on eight rivers (Table 1, Figure 1). Sample sizes ranged from 21 to 97 fish captured. Service personnel took the fish, on ice, back to Deadhorse, Alaska. They were frozen, then shipped to the Service lab in Anchorage. There they were held at -80°C until muscle, liver, eye, and heart tissues were dissected. Long term storage of tissues is also at -80°C.

# Offshore Collections

Our target was 200 fish collected from one location during a five-day period, three times during the summer season. Our sample sizes were determine by availability during that five-day period. Samples were collected from near Prudhoe Bay in the area around Endicott causeway June 24-26, July 29 - August 1, and September 8 -11, 1987 by Envirosphere Company personnel using fyke traps. Service personnel worked at Envirosphere's Deadhorse laboratory to dissect and freeze samples before shipping them back to the Service laboratory in Anchorage.

Samples were collected from the Camden Bay area at Kongaevik Point and Simpson Cove on July 1, 1987 by personnel of the Service's

		UTM coordi	Number		
Population	Grid	Latitude	Longitude	char	Date
Aichilik River	78.1	7600000	410000	40	0/06
Site 2	7W 7W	7699000	419000	40 70	9/00
Anaktuvuk River	50	7626000	565000	40	5/86
Rabbage River	511	,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,	505000		5700
Site 1	7W	7626000	579000	53	8/87
Site 2	7W	7619000	575000	21	8/87
Canoe River	7W	7611000	593000	35	9/86
Canning River					
Site 1	6W			27	5/86
Site 2	6W	7716000	525000	70	8/87
Site 3	6W	7691000	516000	62	8/87
Marsh Fork	6W	7665000	539000	29	5/86
Shublik Spring	6W	7698000	535000	59	8/87
Echooka River	6W	7685000	489000	24	4/86
Egaksrak River	7W	7700000	435000	41	5/86
Firth					
Site 1	7W	7625000	506000	64	8/87
Site 2	7W	7610000	495000	47	8/87
Joe Creek	7W	7646000	501000	40	9/86
Hula Hula River					
Site 1	6W	7741000	614000	15	10/85
Site 2	6W	7712000	609000	37	10/85
Site 3	6W	7692000	598000	59	10/85
Site 4	6W	7735000	613000	97	8/87
Ivishak River	6W	7690000	519000	50	9/86
Kavik River	6W	7690000	519000	40	9/86
Kongakut River					
Site 1	7W	7710000	473000	40	9/86
Site 2	7W	7668000	465000	90	8/87
Lupine River	7W	7659000	439000	48	8/87
Ribdon River	7W	7615000	460000	40	5/86
Sadlerochit Spring	6W	7730000	595000	62	8/87

Table 1.- Populations of Arctic char collected from rivers of the North Slope of Alaska and Canada with site location, number of samples collected from each site, and date of collection.



# Figure 1.- Sampling sites for Arctic char genetics research.

Fairbanks Fisheries Assistance Office.

#### Electrophoresis

We used horizontal starch-gel electrophoresis to identify protein products of gene loci following the methods described by Utter et al. (1974). Buffers and staining procedures were after Allendorf et al. (1977), and isozyme nomenclature was that of Allendorf et al. (1983). Gel buffers included: AC (Clayton and Tretiak 1972) pH 6.1, 6.8; AC+ (AC plus 30 mg NAD); RW (Ridgway et al. 1970) pH 8.2; EBT (Boyer et al. 1963) pH 8.5.

Building on our previous work (Everett and Wilmot 1987), statistical results are based on successful resolution of 42 loci coding for 19 enzymes in three tissues (Table 2). The loci we used are those with nearly complete data sets and consistent results, including good resolution and a repeatable pattern of expression.

Inferences were made regarding enzyme expression based on: 1) assumptions of parallel expression with that of other salmonids with experimentally determined patterns of inheritance (especially Johnson 1984), 2) comparisons based on different tissue expression, and 3) the known molecular subunit structure of the enzymes. Mobilities of enzymes were measured relative to the common electrophoretic phenotype observed in samples of Anaktuvuk River Arctic char. Table 2.- Enzymes, Enzyme Commission (E.C.) numbers<sup>a</sup>, and loci examined in samples of Arctic char collected from northern Alaska in 1987. Buffers include: AC (Clayton and Tretiak 1972), pH 6.1 and pH 6.8; AC+ = AC + NAD; RW (Ridgway et al. 1970), pH 8.2; and EBT (buffer of Boyer et al. 1963, modified by Washington Department of Fisheries biologists), pH 8.5. Tissues include muscle (M), liver (L), and eye (E). The pairs of loci listed in parentheses are electrophoretically indistinguishable (isoloci; Allendorf and Thorgaard 1984). For these analyses, each member of a locus pair was treated as an individual locus with variation assigned to one of the two loci.

Enzyme	E.C. number	Loci	Buffer	Tissue
Adenylate kinase	2.7.4.3	Adk1	AC 6.8	М
Alcohol dehydrogenase	1.1.1.1	Adh1	RW	L
Aconitate hydratase	4.2.1.3	Aco4	AC 6.8	L
Aspartate aminotransferase	2.6.1.1	Aat1,2 .	RW	L,E
•		Aat(3,4)	RW	M
Creatine kinase	2.7.3.2	Ck1,2	RW	M
		Ck3	RW	Е
Fumarate hydratase	4.2.1.2	Fh1 <sup>b</sup>	AC 6.8	M
ß-Ga	3.2.1.30	Hex1 <sup>b</sup>	AC 6.8	L
Glucose phosphate isomerase	5.3.1.9	Gpi(1,2)	RW	M
		Gpi3	RW	M
Glutathione reductase	1.6.4.2	Gr1	RW	L
Glyceraldehyde-3-phosphate				
dehydrogenase	1.2.1.12	Gap3,4	AC 6.1	Е
Glycerol-3-phosphate				
dehvdrogenase	1.1.1.8	G3p1,2	AC 6.1,RW	M,L
Glycyl-leucine peptidase	3.4.11.	PepGL1	EBT	M
Isocitrate dehydrogenase	1.1.1.42	Idh1,2	AC 6.8	M
		Idh(3,4)	AC 6.1,6.8	L,E
Lactate dehydrogenase	1.1.1.27	Ldh1,2	RW	M
		Ldh3,4,5	RW	Е
Levent strent streigs		Ldh	RW	L
Leucy1-g1ycy1-g1ycine	2 / 12	Bont CC1	FDT	м
peptidase	3.4.13.	Mab(1 2)		M
Malate denydrogenase	1.1.1.5/	Mah(2, k)	AC 6 1	M
Malate denydrogenase	1 1 1 40	Mdbol 2 2	AC 6.1	M
(NADP-dependent)	2 7 5 1	Pom1 2	DW DW	M
rnospnoglucomutase	2.7.3.1	$P_{cm}(2, h)$		TM
6-Phosphogluconate		rgm(3,4)	AC 0.1	11911
dehydrogenase	1.1.1.44	6Pg1	AC 6.8	M.L.E
Sorbitol (iditol)		0		
dehydrogenase	1.1.1.14	Sdh(1.2)	RW	L
Superoxide dismutase	1.15.1.1	Sod1	RW	L
Xanthine dehydrogenase	1.2.3.2	Xdh1 <sup>b</sup>	RW	L

<sup>a</sup> Enzyme Commission, IUCBN.

(citation).

<sup>b</sup> Variation was observed at these loci but poor resolution in many samples prevented inclusion in the analyses.

DRAFT

#### STATISTICAL METHODS

#### Amount of Genetic Variation

The amount of genetic variation is estimated by determining the percent of loci that are polymorphic (P), and the mean percent of heterozygous loci per individual (H). Expected average heterozygosity for each locus is calculated using allele frequencies of observed genotypes in each population and expected random mating (Hardy-Weinberg) proportions:

$$H = 1 - \left(\sum_{j=1}^{L} \sum_{i=1}^{A_{j}} P_{ij}^{2}\right) / L$$

where L is the number of loci,  $A_j$  is the number of alleles at the jth locus, and  $P_{ij}$  is the frequency of the ith allele at the jth locus.

The standard criteria for polymorphism (P) is the percent of the loci examined in a population in which the frequency of the common allele is less than or equal to 0.99.

For this and subsequent analyses, isoloci (duplicated locus pairs with indistinguishable mobilities) were counted as two individual loci and all observed variation was attributed arbitrarily to only one locus of the pair.

#### Genotypic Distribution

Observed genotypes in samples were tested for conformance to random mating (Hardy-Weinberg) proportions. A chi-square test was used to determine whether the frequency of genotypes for each locus equal those expected from calculations of probable combinations of alleles (with the frequencies we observed) joining at random. For each population sampled, a multiple simultaneous chi-square test was done by summing the chi-square values over all the variable loci, summing the degrees of freedom, and comparing these values to a chi-square distribution.

13

#### Genetic Heterogeneity

Criteria for joining collections is based on a joint resolution by the West Coast interagency working group doing genetic stock identification of Pacific salmon. It has been agreed that the criteria for joining collections from the same drainage is a probability greater than 0.05 that the collections -- made at different sites or in different years -- are not significantly different using a chi-square or log likelihood ratio (G-test) statistic.

Collections from different geographic areas are not joined before basic analyses. After estimates of percent composition are made, populations can be joined to reflect biological and/or management groups, and the bootstrap variances are recalculated.

To test the heterogeneity between paired populations, we used multiple simultaneous G-tests (Sokal and Rohlf 1981). G-tests were performed for each locus, and G-values and degrees of freedom for each locus were summed over all loci in all pairs and tested against a

14

DRAFT

chi-square distribution. Because of the robustness of the test, only cells with expected values less than 1.0 were combined (Felsenstein 1967).

When making all possible pairwise comparisons for an overall G-test, the large number of non-independent pairwise comparisons (120) makes it possible that a percentage of the comparisons could appear significantly different by chance. Consequently, the probability value required to demonstrate a significant difference among all collections was modified for this analysis according to Cooper (1968) to eliminate spurious correlations. This involves increasing the criteria for a significant test to reflect the number of tests, such that a probability of 0.05 would be divided by the number of tests (here 120). It would then be necessary to get a chi-square value from the table that corresponded to a probability value less than 0.0004 for a comparison to be considered significantly different.

#### Genetic Similarity

The genetic similarity among baseline collections was determined using computer programs by Donald Campton of University of Florida. The program calculates Nei's index of genetic identity (1972; 1978) using the probability of identity of gene pairs between populations averaged over all loci. We report the results of the analyses that compensate for the unequal sample sizes of the collections (Nei 1978).

The normalized identity of genes between two populations, X and Y, is defined as:

# $I = J_{XY} / SQRT (J_X J_Y);$

where  $J_X$ ,  $J_Y$ , and  $J_{XY}$  are the arithmetic means over all loci of the probabilities of identity between gene pairs among populations.

Identity values are scaled from 0.0 to 1.0; 0.0 corresponds to complete allele substitution at all loci, and 1.0 to populations that are electrophoretically indistinguishable at all loci studied. Genetic distance is calculated as the negative natural log of the identity value.

Genetic identity values were used in a clustering algorithm (UPGMA: Sneath and Sokal 1973) modified by Donald Campton to produce a dendrogram of relationships among populations. The average linkage method of clustering was used, and this analysis was also weighted to reflect unequal sample sizes.

#### Gene Diversity Analysis

Gene diversity analysis determines the source of observed variation, i.e., what proportion of the observed variation is between individuals within populations, as opposed to differences among populations or groups of populations. Our analysis was done with a computer program by Donald Campton based on the work of Nei (1973) and Chakraborty (1980). Modifications include use of simple unweighted arithmetic averages of population samples within sites rather than weighting gene frequencies within sites by the number of samples.

Sample data were analyzed by individual subpopulations (sites), by

15

subpopulations of different drainages, and by the combined populations of a region. The combined total amount of genetic variation of all populations studied was partitioned into within- and betweensubpopulation diversity components. The total gene diversity ( $H_T$ ) over all subpopulations equals the average heterozygosity within the subpopulations ( $H_S$ ) plus the average gene diversity between subpopulations ( $D_{ST}$ ). The diversity between subpopulations ( $D_{ST}$ ) can be broken down to differences between sites within a drainage ( $D_{BS}$ ) and differences between populations of different drainages ( $D_{BD}$ ). The relative magnitude of gene differentiation among populations ( $G_{ST}$ ) was estimated as  $D_{ST}$  /  $H_T$  or ( $D_{BS}$  +  $D_{BD}$ ) /  $H_T$ , and can be expressed as a percentage.

#### Genetic Stock Identification

Genetic stock identification was done using a maximum-likelihood estimate program modified by Sam Nelson and Jerome Pella (National Marine Fisheries Service, Auke Bay, Alaska) with the expectation maximization (EM) algorithim as used by Milner et al. (1983), and refined by Millar (1987). Estimates and standard errors are obtained using a bootstrap technique (Demster 19??, Efron 1982) whereby the mixture is resampled 100 to 200 times.

#### Simulations

We used genetic stock identification methods four ways to evaluate

16

11/29/88

their effectiveness for North Slope Arctic char. First, we constructed an artificial mixed stock by combining all baseline data. The baseline data characterizes each fish stock; it is used for a reference by which the artificial mixed stock fish are assigned as a percent composition to the stock-of-origin. The program gives point estimates and variances of the estimates for a sample of known composition. The actual contribution of each populations to the artificial mixture is determined by sample size. We are then able to compare estimated percent composition of the artificial mixed stock to actual expected values using a chi-square statistic using number of fish estimated to have come from each stock, compared to number of fish expected (actual mixture composition).

17

in the second second

Second, we did incremental mixed-stock simulations where each baseline population was added to an artificial mixture at 20% increments, with baseline data from other populations used to total 100%. This tests the accuracy and precision of the estimates for each stock over the range of possible contributions from 0 to 100%. Point estimates and standard errors are graphed with actual contributions in known estimates for comparison.

Third, we did simulations where each baseline stock, one at a time, was used to make up an artificial mixed stock. When tested against the baseline made up of all stocks, these simulations indicate how the fish are allocated, and to which stocks incorrectly allocated fish are assigned.

Endicott Samples

We also did estimates of the percent composition of mixed fishery samples taken from Prudhoe Bay adjacent to the Endicott Causeway in June, July, and August of 1987. We used baseline data from 14 Arctic char populations and bootstrap resamplings to determine mean estimate and the error around the mean of the estimates.

Populations with an estimated stock contribution of 1% or less are not used. They are considered spurious, as the maximum likelihood analysis program always assigns some fish to each baseline specified as a possible contributor.

#### RESULTS

We have information on population parameters compared among baseline populations, computer simulations on known samples for accuracy and precision, and mixed-fishery composition estimates for Endicott samples.

#### 1987 Collections

Most Arctic char captured are juveniles, with some small resident adults. A small sample of char was collected upstream from the waterfall on the Babbage River (site number 2) for comparison of these resident fish with anadromous populations. Firth River sampling site number 2 is in Alaska, and was included for comparison of upstream and downstream populations in this drainage. Upstream Firth fish were

DRAFT

small, and many appeared to have mature gonads.

No Arctic char were captured at sites that were sampled near the Chukchi Sea, including the Singaruak, Walakpa, and Kugrua rivers. No Arctic char were captured at the Shaviovik River, the downstream end of the Colville River, or the Anaktuvuk, which is tributary to the Colville.

Approximately 232 Arctic char were collected from Prudhoe Bay in late June, 137 in late July, and 166 in early September. From the Camden Bay area, 50 char were collected near Kongaevik Point and 50 from Simpson Cove.

#### Amount of Genetic Variation

Of the 46 gene loci we examined, 15 are variable and were used in the analysis. Allele frequencies were calculated for each population, and relative mobility of allelic variants were measured (Appendix A). Four additional enzyme loci (<u>Hex1</u>, <u>FH1</u>, <u>Pgm3,4</u>, and <u>Xdh1</u>) are known to be variable in these Arctic char. <u>Aat1</u>, <u>Hex1</u>, and <u>Xdh1</u> were scored in 1986, when we had live fish to sample in the lab. However, these gene loci are particularly sensitive to sample handling and storage, and consistently good resolution could not be obtained in 1987 when samples were brought from the field frozen. The other 27 loci studied are monomorphic in all populations.

Percent of loci polymorphic (P) and average heterozygosity per locus (H) for the 16 populations of Arctic char sampled were calculated for combined 1986 and 1987 collections (Table 3). The

19

Table 3.- Expected average percent of fish heterozygous per locus (H), and percent of loci examined that were polymorphic (P) in 16 populations of Arctic char sampled from rivers of the North Slope of Alaska in 1986 and 1987. Samples in some drainages were combined if G-test p > 0.05. The average value of H is weighted by sample size. Heterozygosities were recalculated in 1988 to include only loci that were studied in both years. Hex, XXO, and FH were excluded.

		Max		
Drainage / Sites	Year	N	% H	% P
Aichilik River	1986/1987	85	5.04	23.81
Colville River				
Anaktuvuk	1986	40	3.81	19.05
Babbage River				•
Canoe River	1986	35	2.51	9.76
Site #1	1987	53	2.48	11.90
Site #2	1987	21	2.43	7.14
Canning River				
5 Sites	1986/1987	212	4.13	26.19
Egaksrak River	1986	41	4.32	21.43
Firth River				
3 Sites	1986/1987	132	4.29	28.57
Hula Hula River				
Site #1	1986/1987	95	4.57	23.81
Site #2	1986	54	4.66	22.50
Kavik River	1986	40	3.14	14.29
Kongakut River				
2 Sites	1986/1987	85	4.54	21.40
Sadlerochit River	1987	45	1.63	7.14
Sagavanirktok River				
Ivishak/Echooka	1986	74	3.62	26.19
Lupine	1987	45	4.36	19.05
Ribdon	1986	40	5.16	21.95
Average		1097	$3.79 \pm 1.02$	$19.01 \pm 6.68$

Sall and Shares

11/29/88

values of P range from 7.1 to 35.7% (average: 19%). The average population heterozygosity ranges from 1.6 to 5.2% for the combined 1986 and 1987 collections, and the weighted average heterozygosity per individual over all populations is 3.8%.

21

and the second second

#### Genotypic Distributions

Significant deviation from expected values can indicate non-random mating, unequal fertility among parents, unequal viability among offspring (selection), migration from other populations, or failure to collect a random sample from the population. Within the collections of Arctic char we studied, individual loci were occasionally out of equilibrium. However, when all loci for a population were considered, there was no evidence of departure from the expected genotypic distributions in any population. The parental generations have apparently been mating at random (no more than one population was detected in any sample), and the collections appear to represent random samples of the populations.

#### Genetic Heterogeneity

Heterogeneity tests between North Slope Arctic char populations were done first between collections from the same drainage at different sites and in different years. The data for the differing sites and years of collection within the Aichilik, Canning, Firth, and Kongakut Rivers char subsamples were combined (Appendix A) as they are

11/29/88

not detectably different from each other (Table 4). The char collected from the upstream sites (number 2 and number 3) of the Hula Hula River are not distinct genetically, and were combined as site number 2. Samples from Hula Hula site number 1 in 1986 and 1987 are not significantly different, and were combined as site number 1. The five sites in the Canning River Drainage include char from the Marsh Fork and Shuklik Springs.

22

The three collections from the Babbage River Drainage (from the Canoe River, from a site downstream from the waterfall, and from a site above the waterfall) could not be combined because they are significantly differentgenetically. The populations of the major tributaries of the Sagavanirktok River are significantly genetically distinct, though the collection from the Ivishak River is not distinct from that of its tributary, the Echooka. This results in a total of 16 baseline populations for all analyses excepting genetic stock identification procedures, where a baseline of 14 stocks is used.

When data from the 16 collections are compared, most of 120 pairwise comparisons indicate significant genetic differences (p < 0.01) among North Slope char populations when corrected for the number of non-independent tests (Table 5). A summary G-test, including all populations and all 15 variable loci, shows that the Arctic char studied are highly different from each other (G = 1237 with 143 degrees of freedom; p << 0.001).

Genetic Similarity

Same and the state of the second states of the

Table 4.- Log likelihood ratio (G-tests) heterogenity tests among populations sampled at different sites and/or different years. Probability of P>0.05 was used as criteria for combination of samples within a drainage. Degrees of freedom (df) reflects the number of loci in the comparisons.

Population	Year	Sites	G	df	P
Aichilik	1986/1987	2	12.45	11	0.330
Babbage	1986/1987	3	46.95	7	<0.001
Canning	1986/1987	5	19.31	8	0.132
Ivishak	1986	2	11.99	8	0.152
Firth	1986/1987	3	31.64	23	0.108
Hula Hula	1986/1987	2	29.41	9	<0.001
Kongakut	1986/1987	2	5.71	10	0.839

Table 5.- Matrix of genetic heterogeneity, tested pairwise among Arctic char populations of the North Slope, Alaska and Canada. G-values, with degrees of freedom are in parentheses. The significance level was modified according to Cooper (1968) to compensate for the number of pairwise tests (120).

1	Canoe 86	x															
-	Canoc ou	**															
2	Babbage 87-1	19.4	X														
3	Babbage 87-2	38.8	17.0	x													
		(3)	(4)														
4	Firth	110.7	96.1	60.7	X												
		(11)	(12)	(9)													
5	Kongakut	100.0	106.9	70.9	54.4	X											
		(10)	(10)	(9)	(13)												
6	Egaksrak	46.0	102.6	73.1	97.6	45.6	X										
		(8)	(8)	(8)	(14)	(11)											
7	Aichilik	49.1	136.5	92.5	135.5	75.0	33.1	X									
		(10)	(11)	(10)	(14)	(13)	(11)										
8	Hula 1	81.7	150.7	87.7	139.6	46.6	25.7	73.5	X								
		(9)	(11)	(8)	(14)	(12)	(13)	(14)									
9	Hula 2	99.8	154.4	91.8	128.2	59.6	29.5	97.5	29.4	X							
		(8)	(9)	(8)	(10)	(10)	(9)	(10)	(9)								
10	Sadlerochit	99.2	75.4	66.6	164.4	180.8	1/9.6	212.5	247.2	226.0	X						
		(4)	(5)	(5)	(11)	(10)	(/)	(11)	(10)	(8)							
11	Canning	8/./	129.6	63.8	125.9	/5.0	43.2	12.9	59.8	100.7	208.2	X					
		(10)	(10)	(9)	(14)	(13)	(12)	(14)	(13)	(9)	(10)	07 0	v				
12	Kavik	124.3	60.4	32.6	/8.4	61.8	/0.4	124.2	08.5	50.2	132.0	(10)	•				
		(6)	(6)	(5)	(10)	(10)	(9)	(11)	(9)	70 7	(0)	(10)	20.2	v			
13	Ivishak	134.4	12.3	38.9	(12)	40.2	(10)	(12)	(11)	(10)	(9)	(12)	(10)	•			
	B 11 1	(9)	(9)	(/)	(13)	(11)	(10)	(12)	(11)	(10)	156 7	112 /	(10)	51 5	v		
14	Ribdon	89.3	82.8	(10)	(11)	40.3	(11)	(12)	(12)	(10)	(0)	(12)	(10)	(11)	А		
1.5	T	(10)		(10)	(11)	20 2	20 0	(13)	56 6	74 7	141 7	66 1	79 0	51 1	26 0	x	
12	Lupine	22.4	(7)	(7)	(12)	(12)	(9)	(12)	(11)	(0)	(7)	(11)	(8)	(10)	(10)		
10	A 1- 4	(0)	(/)	62 0	(13)	70 0	75 /	103 /	85 2	128 8	108 8	29 5	88.5	57.2	96.7	64.2	x
10	Anakcuvuk	(10)	(0)	(0)	(13)	(12)	(13)	(13)	(13)	(12)	(8)	(11)	(10)	(11)	(13)	(10)	**
		(10)	(9)	())	(13)	(12)	(13)	(13)	(13)	(12)	10	11	10	10	11	15	16
		1	2	3	4	5	6	1	8	9	10	11	12	13	14	15	10

No allele substitutions were observed at any locus. Genetic identities are high between North Slope char populations. All pairwise comparisons have values greater than or equal to 0.981, corresponding to a genetic distance of 0.019. The Sadlerochit River population is responsible for the lowest identity values among the char studied. Without this unique population, the identity values among anadromous North Slope char are 0.989 or higher, ranging up to complete identity, 1.000 (Table 6).

A dendrogram (Figure 2) illustrates the genetic relationships among Arctic char populations of tributaries of the Beaufort Sea. Again, the Sadlerochit River char are the most unlike the other populations.

#### Gene Diversity Analysis

The absolute and relative magnitude of the diversity among subpopulations was analyzed hierarchically (Table 7). Approximately 8% of the observed variation is due to differences among the populations of the 11 drainages sampled. Less than 1% is due to differences among populations of different sampling sites within drainages. Variation among individuals within populations accounts for 91.1% of the total gene diversity.

#### Genetic Stock Identification

Artificial Mixed Stock Simulations .- For these genetic stock

Table 6.- Matrix of Nei's (1978) gene identity values pairwise among 16 populations of Arctic char sampled from the North Slope of Alaska rivers in 1986 and 1987.

	Population																
1	Canoe 86	1.000															
2	Babbage 87-1	0.998	1.000														
3	Babbage 87-2	0.995	0.999	1.000													
4	Firth	0.999	0.998	0.996	1.000												
5	Kongakut	0.999	0.997	0.996	0.999	1.000											
6	Egaksrak	0.998	0.994	0.994	0.996	0.999	1.000										
7	Aichilik	0.999	0.996	0.995	0.998	1.000	0.999	1.000									
8	Hula 1	0.998	0.995	0.995	0.996	0.999	1.000	1.000	1.000								
9	Hula 2	0.997	0.989	0.991	0.991	0.996	0.999	0.997	0.998	1.000							
10	Sadlerochit	0.989	0.996	0.996	0.993	0.990	0.985	0.989	0.986	0.981	1.000						
11	Canning	0.997	0.998	0.997	0.999	0.999	0.998	0.999	0.999	0.995	0.991	1.000					
12	Kavik	0.996	0.998	0.999	0.997	0.998	0.998	0.998	0.998	0.996	0.992	0.999	1.000				
13	Iviskak	0.997	0.998	0.998	0.998	0.999	0.998	0.999	0.999	0.994	0.991	1.000	0.999	1,000			
14	Ribdon	1.000	0.997	0.995	0.999	1.000	0.998	0.999	0.998	0.995	0.989	0.998	0.997	0.998	1.000		
15	Lupine	0.999	0.997	0.995	0.999	0.999	0.997	0.999	0.997	0.993	0.991	0.998	0.996	0.997	1.000	1.000	
16	Anaktuvuk	0.996	0.998	0.997	0.999	0.998	0.995	0.997	0.996	0.990	0.993	0.999	0.998	0.999	0.996	0.997	1.000
		1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16

DRAFT



# **GENETIC SIMILARITY**

Figure 2.- Relationships based on Nei's (1978) index of genetic identity of 16 populations of Arctic char from rivers of the North Slope of Alaska and Canada.

			Absolute ge	ne diversity	Relative diversity (%)			
Drainage	# of sites	Within sites	Between sites	Between drainages	Total	Within sites	Between sites	Between drainages
Babbage River	3	0.0256	0.0020		0.0276	92.9	7.1	
Hula Hula River	2	0.0463	0.0009		0.0472	98.1	1.9	
Sagavanirktok	3	0.0429	0.0013		0.0442	97.1	2.9	
Average	16	0.0383	0.0004	0.0033	0.0420	91.1	0.9	8.0

Table 7.- Gene diversity analysis (Nei 1973; Chakraborty 1980) among populations of Arctic char from rivers of the North Slope of Alaska and Canada. The average values represent data from all 16 populations from the 11 drainages studied in 1986 and 1987.
identification analyses, data from 14 stocks were used as a baseline. The Sadlerochit char were excluded from the analyses, as they are not anadromous. Fish from Babbage River upstream of the waterfall were also excluded from the GSI analyses as they probably contribute little to migratory stocks.

29

<u>Artificial Mixture</u>.- We used the data from baseline spawning populations to make an artificial mixture (N = 1032) of known composition. The actual values for 7 of 14 populations were within an 80% confidence interval (point estimate ±1.28 standard deviations) of the estimated allocation from GSI (Appendix C and Figure 3).

Canoe, Hula Hula site #2, and Ribdon River populations were estimated at essentially zero while actually present at 3, 5, and 4% respectively. Eighty percent confidence intervals for estimated values included zero for Egaksrak, Lupine, and Anaktuvuk River populations.

Babbage, Egaksrak, Kavik, and Lupine char point allocations are not significantly different than actual values ( $\chi^2$ : p > 0.5). Kongakut and Anaktuvuk River composition estimates are significantly different than actual composition ( $\chi^2$ : P < 0.05), but if corrections are made for the number of non-independent tests (Cooper 1968) are not. The actual contributions to the artificial mixed stock of the populations listed above are within the 80% confidence intervals around the estimated values. Only the actual contributions of Canoe, Hula Hula site #2, and Ribdon River fish (erroneously estimated at zero contribution to the mixture) and Canning River fish are not

DRAFT

and the state of the second has a state of the



Figure 3.- Maximum likelihood method of genetic stock identification on an artificial mixed stock of North Slope Arctic char. Estimated numbers of fish ( $\Box$ ) are given with 1.28 standard deviations calculated from bootstrap resampling. Actual values ( $\blacklozenge$ ) are numbers of fish from each stock in the artificial mixture.

DRAFT

within two standard deviations (95% confidence interval) of their estimates.

Incremental Simulations.- Incremental artificial mixed stock simulations showed that some populations of char are identifiable within a whole range of contributions to a simulated mixed fishery (Appendix D and Figures 4a-n). Data for 14 populations was used to identify, sequentially, each population as 100% of an artificial mixture. In the simulations, each population studied was identified as making up 78% or more of its mixture. Eight of 14 stocks were greater than 90% correct, and three were greater than 95% correct.

Fourteen percent of the Babbage Drainage Canoe River fish were incorrectly allocated to the Babbage mainstem population. Over 12% of Kongakut fish were incorrectly allocated to Aichilik and Egaksrak River, and conversely, 16.5% of Aichilik fish were incorrectly allocated to Kongakut and Egaksrak. Eight percent of Hula Hula site #1 fish were allocated to Egaksrak, as were 7% of Hula Hula site #2 fish. Twelve percent of Canning fish were incorrectly allocated, as were 16% of the Ribdon char. Twelve percent of Ribdon fish were allocated to Lupine River, of the same drainage. The Anaktuvuk population was properly allocated 98% of its fish by the GSI technique.

<u>Twenty Percent Incremental</u>.- When different populations were used to make up different percentages of mixture files, in 20% increments from 0 to 100%, several populations were correctly allocated throughout the range of the simulations (Figures 5a-n). These were



DRAFT



Figure 4.- Incremental artificial mixed stock simulations for each of 14 populations of North Slope Arctic char. Estimated using maximum likelihood techniques with 200 bootstrap resamplings.





Figure 4.- Continued.



DRAFT



Figure 4.- Continued.



Figure 5.- Incremental artificial mixed stock simulations for each of 14 populations of North Slope Arctic char. Contributions of each population, in 20% increments, are estimated using maximum likelihood techniques with 200 bootstrap resamplings.

35

and a second second



# Figure 5.- Continued.

ESTIMATED PROPORTION

19

1.0 -

0.8

0.6

0.4

0.2

0.0

0.0

0.2

0.4 0.6 TRUE PROPORTION

0.8



1.0



Figure 5.- Continued.

37

and a second the second and the second se

1 and a fill of the share of the second state of the second state of the second

DRAFT

Babbage site #2 and Sadlerochit River char (neither of which were used in the GSI analyses); Kongakut, Egaksrak, Aichilik, Hula Hula site #1, Ivishak, Lupine, and Anaktuvuk. Canoe River, Firth, Canning, Hula Hula site #2, and Kavik were consistently underestimated, and Babbage site #1 and Ribdon were very poorly "recognized."

<u>Mixed Fishery Samples -- Endicott</u>.- Estimates of the stock composition of mixed Arctic char taken near Endicott Causeway in June (N = 208), July (N = 126), and August 1987 (N = 166), were made using maximum likelihood estimation, bootstrap resampling, and the 14-stock baseline already described. Eight percent confidence intervals were generated around the point estimates for each population (estimate ±1.28 standard deviations); and for each month (tabulated in Appendices E, F, G, and graphed as Figures 6, 7, and 8).

For June, only Ivishak, Lupine, and Anaktuvuk estimates did not include zero in the 80% confidence interval. When the contribution of the Sagavanirktok fish (Ivishak, Lupine, and Ribdon) are summed and the variances recalculated, they contribute 66%  $\pm 20.9\%$ . Anaktuvuk contributes an additional 14%  $\pm 10.9\%$ , together accounting for 80% of the fish sampled.

In July, four populations made contributions where zero was not within the 80% confidence interval. Hula Hula site #1 (12%  $\pm$ 10.2%) and Kavik (10%  $\pm$ 8.3%) drop out of the estimates if a 95% confidence interval is used, as zero would be included in that interval. When the estimates of the three Sagavanirktok tributaries are allocated, then summed and the variances recalculated, the contribution is 42%



Estimates

Figure 6.- Estimated composition (±1.28 standard deviation) of a mixed fishery sample from Prudhoe Bay, Alaska, collected in June 1987. Estimates are made using maximum likelihood techniques with 200 bootstrap resamplings and a 14-stock genetic baseline.

DRAFT



Estimate

Figure 7.- Estimated composition (±1.28 standard deviation) of a mixed fishery sample from Prudhoe Bay, Alaska, collected in July 1987. Estimates are made using maximum likelihood techniques with 200 bootstrap resamplings and a 14-stock genetic baseline.

Anaktuvuk

The she had a second with the subscription of the



Figure 8.- Estimated composition (±1.28 standard deviation) of a mixed fishery sample from Prudhoe Bay, Alaska, collected in August 1987. Estimates are made using maximum likelihood techniques with 200 bootstrap resamplings and a 14-stock genetic baseline.

DRAFT

 $\pm 17.3\%$ . Anaktuvuk contributes 21%  $\pm 8.5\%$ . Sagavanirktok and Anaktuvuk fish, then, account for 63% of the fish sampled in July.

43

In the August sample, all three Sagavanirktok tributary stocks are represented. When the allocations of Ivishak, Ribdon, and Lupine fish (43%, 11%, and 24%, respectively) are summed and the variance recalculated, they equal 78% ±13.3% (80% confidence interval: 0.6078 - 0.9472; 95% confidence interval: 0.5176 - 1.0375).

### DISCUSSION

In 1987 additional samples of Arctic char were collected for baseline genetic data, including samples from populations with non-adadromous, small adults. We did simulations using the maximum likelihood estimation program to test the accuracy and precision of our combined baseline data from 1986 and 1987 collections. Using genetic stock identification techniques, the compositions of collections of mixed stocks of char from near Endicott Causeway at the mouth of the Sagavanirktok River was estimated for June, July, and August 1987. With this information, we are able to discuss population characteristics of North Slope Arctic char, the quality and potential of our current baseline data, and the possible implications of the estimates of Endicott char stock composition, plus direction and capabilities for further study.

### <u>Collections</u>

Our genetic baseline for Arctic char covers the major migratory stocks from the McKenzie River of Canada to Point Barrow in Alaska. Our mixed stock samples are limited to the area around Endicott Causeway, at the mouth of the Sagavanirktok River, Alaska. Collections of char taken near Barter Island are too small to be useful.

44

Sampling additional char necessitated modifying our 1986 baseline data sets, and the enzyme protocol we used. In 1988 we were more conservative in the number of loci we considered to be reliably scoreable. Many of the char samples we obtained in 1986 were brought back alive, and consequently were of very high quality when analyzed electrophoretically. Though every effort was made in 1987 to bring back quality samples from the field, frozen samples are not likely to be as good. In 1987 we were unable to consistently score two highly variable loci, <u>Hexl</u> and <u>Xdhl</u>.

Fish from more sites in freshwater tributaries; second samples from same or similar sites; and larger sample sizes were collected. No Arctic char were found in three rivers draining to the Chukchi Sea in August 1987, or in certain other rivers of the Beaufort Sea area. We did collect char representing different forms associated with non-migratory life history.

### Amount of Genetic Variation

The amount of genetic variation in North Slope Arctic char is typical of fish species in general (Nevo 1978), and slightly higher

than average among salmonids that have been studied (Utter et al. 1981). Our overall estimate of variability, measured as average heterozygosity, for char from our combined 1986 and 1987 collections is slightly lower than that for 1986 alone. Two loci were excluded from the 1987 analyses, and samples from populations from small, isolated populations in springs and from upstream in the Babbage and Firth River Drainages were included.

45

#### Combining Baseline Data

With the exception of the Babbage, Hula Hula, and Sagavanirktok River systems, we found that collections of Arctic char made in different years and in different parts of the same drainage are not significantly different in allele frequencies (G-test: p < 0.01) and could be combined. Multiple samples taken in 1986 and 1987 from the different sites within the Canning, Firth, Aichilik, Kongakut, and Ivishak are not significantly different in allele frequencies. It is best to combine baseline information for stocks that are both genetically and geographically similar.

The amount of precision in genetic stock identification estimates has been shown to be consistent with the level of divergence among stocks (Milner et al. 1981). Collections of fish that have not diverged significantly should be combined, particularly since larger numbers of baselines result in smaller percentage contributions allocated to a greater number of stocks. Smaller estimates typically have relatively large errors (comparable as coefficients of

variation). Milner et al. (1986) found that stock composition estimates of less than 5% for a given population generally are poor. With many baselines, it is likely that more stocks will be identified in smaller percentages than if more baselines are used.

46

Though we sometimes find genetically similar stocks from different drainages or even regions, they should not be combined before analyses. Geographically remote stocks should be combined after the fact because we know that the EM algorithm is unable to discriminate between them, and will assign fish to one or both of them. Wood et al. (1987) have done simulations showing that estimates should be made for individual stocks that are genetically similar but unrelated geographically. These estimates can be pooled, and the variances recalculated. Recalculated variances are smaller than the sum of the individual variances.

Collections from drainages supporting both resident and migratory forms of char can apparently be combined in the case of Firth and of Canning River char. We found no evidence that these life history forms represent separate populations. If genetic differences were detected between resident and migratory forms it could be due either to separate evolutionary lines or recent reproductive isolation. Recent divergence may be due to behavioral or physical isolation, which allows genetic differences to accumulate. It is certainly possible that similarities among geographically isolated groups may be due to selection, founding events, or by chance convergence of electrophoretic phenotypes at structural loci.

In other salmonid populations that have been studied, e.g.,

rainbow trout (Allendorf and Utter 1979) and brown trout (Ryman and Stahl 1981), only a small percentage of the divergence among populations is due to the ecological distinction between resident and migratory forms. Resident populations of North Slope Arctic char could either be composed of a separately evolved group with physiological or behavioral isolating mechanisms from migratory groups, or they could have arisen independently in various drainages where condition made it unfavorable or impossible to migrate.

47

With the exception of the Babbage River, the Arctic char populations of a given drainage had no detectable statistical difference between fish known to include residents (small adults) and juveniles from the main range of the anadromous adults. Within collections with both dwarf adults and juveniles of unknown life history propensities towards anadromy, we found no evidence of disequilibrium of allele frequencies that might be expected if more than one ecologically distinct breeding population was included in one collection.

The fish collected from the Sadlerochit Springs area are thought to be entirely non-migratory (Craig 1977), and were consequently excluded from mixed-stock fisheries analyses. They are genetically distinct in allele frequencies from other North Slope char populations, though there are no major differences such as allele substitutions. This distinction from other populations could be easily explained by isolation in a closed system, and periodic episodes of low effective population size. They are genetically distinct, but may well have become so by losing genetic variation.

DRAFT

Loss of genetic variation is often a symptom of low population size and random genetic drift.

Thus, resident groups could either resemble each other across the North Slope, or could most closely resemble the migratory groups in their drainage, with local divergence due to selection or genetic drift (random changes) in presumably small populations. We found that residents generally resembled those populations "expected" to be anadromous within the drainage, or in the case of the physically isolated Sadlerochit Springs char, to be quite different.

## Genetic Differences Among Populations Within Drainages

We studied three collections of char from the Babbage Drainage. Those of the Canoe River, those from below the waterfall, and those from above the waterfall have all apparently diverged. The distinction may be less one of Taxonomy than relatively recent reproductive isolation and genetic drift. Bain (1974) has observed small adults spawning with anadromous char below the falls. While the downstream group may have emigrants coming in, the group above has no migration in. If condistions were unfavorable upstream and the population size became severely reduced, a shift in allele frequencies could occur due to random processes without strong directional selective forces being responsible.

The population sampled downstream of the Babbage Falls probably receives at least one successful migrant per generation, enough to prevent species divergence (Allendorf and Phelps 1980). The upstream

group meanwhile is probably diverging in allele <u>frequencies</u> due to a small effective population size. Though divergence between char of these sites on the Babbage has been observed, genetic similarities between them still results in their closeness on a dendrogram showing genetic relationships among populations.

49

The distinctness of the Canoe River population, from a tributary to the Babbage River, may relate to the small sample size (N = 21) available from that population. Sampling error could have resulted in the inclusion of a non-random sample, also indicated by disequilibrium at the <u>Gpi3</u> locus.

The Sagavanirktok is a large system, with a number of tributaries and considerable char populations. Of the collections we made in 1986 and 1987 from four tributaries, two are genetically similar. The Echooka is tributary to Ivishak River, and Echooka char are not significantly different genetically from the Ivishak fish. The others are distinct, though Lupine and Ribdon are relatively closely related using a genetic similarity index, and simulations show that Ribdon fish are allocated to Lupine.

The Hula Hula River char from two different sites are genetically distinct. Though this river is apparently not complicated by different tributary stocks, it is possible that the two different sites are used preferentially by different char, with spatial and/or temporal variation in spawning. Some reproductive isolation is apparent, though they do cluster together fairly closely on a dendrogram of genetic similarities.

The complicating factor in the discussion of Hula Hula River char

11/29/88

is that Egaksrak River char are even more similar, on average, to Hula Hula River site #1 fish than are Hula Hula River site #2 char. The Egaksrak River is actually between and very close to the Aichilik and Kongakut rivers. The char of the latter rivers are very similar genetically, while those of the Egaksrak most closely resemble those of the Hula Hula River. Though this is not "tidy,", it may well be that the colonizers of the Hula Hula River were from the Egaksrak (or vice versa), or the similarity may be due to chance convergence of allele frequencies, or possibly selection.

50

The relationship between Canning and Ivishak River Drainage char populations also require discussion. From Craig's work (1971) it is apparent that there is migration of Arctic char between the Canning and Ivishak Rivers. Non-spawners were tagged in the Ivishak, and were observed spawning in the Canning. Non-spawners in the Canning were tagged, and were seen in spawning condition in the Ivishak. Though we have no evidence of individual fish spawning in both places, it is interesting that we are currently unable to discriminate genetically between the Ivishak and Canning River char populations we sampled. Though actual interbreeding between char of these populations may be taking place, it is also possible that we sampled actual Ivishak fish from the Canning, or actual Canning fish in the Ivishak. Chance convergence of genotypes is certainly a possible explanation, as is directional selection.

Genetic Divergence Among Populations

After combining collections within drainages, most North Slope Arctic char populations from different drainages are significantly genetically distinct from each other (with the exceptions noted) using a heterogeneity test that emphasizes the effect of variable loci. This information indicates that fish from different drainages are not freely interbreeding, and are most likely true to their spawning streams.

51

Although we have quantified significant genetic differences among populations of char within the Babbage and Sagavanirktok River systems, and among populations of different drainages, the overall genetic similarity among all char studied -- migratory and non-migratory -- is high. On the scale of similarities used (Nei 1972; 1978), both the variable enzymes and the number of enzymes that are consistently monomorphic are considered in evaluating relatedness among stocks. The measured differences among these populations only reflect what is recognized in other taxa as "local" differences (see Ayala and Kiger 1980, or Hartl 1980), relating to fairly recent divergence. We found no fixed differences among populations that would identify them as different taxa. Sadlerochit Springs fish are the most unlike other populations, but this could be explained by loss of genetic variation in a small closed system.

Most of the diversity in North Slope Arctic char is between individuals within subpopulations, and a seemingly small percent is due to differences between subpopulations. Among the char populations we collected in 1986 and 1987, only 8% of the variation is due to statistically detectable differences among fish from different

drainages. This amount, however, is similar to the 7% difference that has been quantified among human racial groups, again indicating current reproductive isolation and considerable difference among stocks.

## Genetic Stock Identification: Simulations

Simulations are particularly needed with North Slope char studies. Other types of information on population dynamics are generally used with GSI to verify or validate what is seen for estimates. Since these other means are not available to address the accuracy and precision of our estimates we must rely on simulations and on limited amount of biological data available. There are currently no large-scale, comprehensive studies of population dynamics, escapements, enhancement, tagging, or scale pattern analyses that include the entire Beaufort Sea area. Certain studies target specific areas, such as Barter Island (Service) or Prudhoe Bay (oil company consultants). Past work did include tagging studies across the North Slope area, but tag returns were limited.

To evaluate the accuracy and precision of our baseline for Arctic char genetic stock identification, a simulation estimating the composition of a known stock was done. Less than half the stocks contributing to this known mixture, composed of baseline data, were allocated to the correct baseline using the GSI technique. Four stocks were estimated at zero contribution.

Poor stock discrimination could be due, in part, to the fact that

this type of estimate is a special case, where all stocks are present in low percentages. Estimates of stock compositon are more precise when less stocks are in higher percentages in a mixture. As large variances around small estimates often include zero, it is not then possible to presume accuracy in near-zero estimates, even though several stocks may be making small positive contributions to a fishery.

53

Given that low percentage estimates are predictably poor, 100% incremental simulations, which use known baseline from an individual stock as a mixture, should represent the best case. Though all baselines tested were at least 78% correct, and one was actually 95% correct, it is apparent from these simulations that certain populations are not well identified using GSI methods. It is probably a factor that the sample sizes for these simulations are small, as several baselines were composed of only forty samples, and some were less.

Incremental simulations do effectively give some indication of where fish are incorrectly allocated by the program. There is some pattern in the way certain char are assigned, e.g., simulations show that some fish are incorrectly assigned to other populations in their own drainage, for instance. The 100% simulation shows that Canning fish that are not allocated to the Canning baseline go to Ivishak and Firth Rivers. In the artificial mixed stock analysis, Canning is underestimated, and Ivishak and Firth stocks are overestimated by a corresponding amount.

The Canning River population allocation might be explained or improved in the future if certain points were considered. In the case

of the Canning River baseline, we combined data from five different sites collected over two different years and including at least two morphotypes representing different life history strategies. Though a test of heterogeneity signified that these collections did not represent significantly different breeding stocks, it is possible that the test was not sufficiently rigorous in defining differences.

54

Another potential problem with the Canning stock identification is indicated by the incremental simulation representing different percentages of contribution by the Canning stock in an artificial mixture. At 20%, which corresponds to the actual percentage of Canning data in the artificial mixed stock simulation, Canning was the most underestimated.

Most important, it is possible that the Canning char stock, as represented by our baseline, is just not sufficiently diverged from other stocks -- especially the Ivishak -- to be recognized by the GSI program. Perhaps it does not have unique genotypes. It is also possible that either our baselines from these two areas have included fish from the wrong stocks, or spawning fish are actually mixing in these two rivers.

In other cases it is not apparent where and why fish are allocated when the estimates and actual values are unlike. In some stocks that are underestimated, the stocks that the simulations show that they should be allocated to are not correspondingly overestimated. Other types of simulations, exploring other cases with different numbers of char artificial stocks, could be designed to eliminate problems we understand or recognize or expect.

## Evaluating Genetic Divergence

adamsto the state of the adams the state of the second state of the

There are different ways of evaluating the degree of genetic divergence among stocks to predict the success with genetic stock identification techniques. It can be done with a scaled index of similarity, such as that of Nei (1972) or with a test of heterogeneity, such as the log likelihood ratio test of Sokal and Rohlf (1981), also called the G-test statistic. Genetic divergence (G<sub>ST</sub>, Nei 1973) also gives an estimate of the divergence among stocks.

We used G-statistics and  $G_{ST}$  to predict that GSI would work to discriminate the composition of mixed stocks of North Slope char. The G-tests indicated that most populations are strongly distinct, even when corrected for the number of independent pairwise tests.  $G_{ST}$ levels are similar to the  $G_{ST}$  levels of salmon species where GSI is used in Washington, D.C., and California. However, Wood et al. (1987) suggest that a modified index of genetic similarity (Nei 1972), or a proportional difference in allele frequencies would be better predictors of successful stock discrimination. Though the G-tests and  $G_{ST}$  are significant, our similarity values are higher than those seen, for instance, in Columbia River chinook.

Milner et al. (1983) evaluated genetic stock identification techniques with actual chinook salmon electrophoretic data. They had baseline, mixed stocks, and other methods, such as escapement counts, coded wire tags, and life history data with which to validate their results. The GSI results are consistent with other types of data. The indices of relatedness, measured as Nei (1972) index of genetic

DRAFT

distance, between chinook population <u>pairs</u> ranged from 0.000 to 0.0737, with an average value of 0.0205. The maximum distance value between North Slope char populations is 0.0192, approximately equal to the average value for chinook. The distance between chinook <u>groups</u> ranged from 0.008 to 0.015, whereas Arctic char distance between groups is approximately 0.003. The <u>within-group</u> variation for chinook ranged from 0.0015 to 0.0382 and averaged 0.0112, compared to 0.04 for North Slope char that were studied. The work of Wood et al. (1987) and comparisons with the chinook data of Milner et al. (198?) suggest that using G-statistics for predicting success in population identification may not be the best method.

## Genetic Stock Identification: Endicott Samples

In the June and August collections, all population contribution estimates where zero was not included in an 80% confidence interval were identified as Sagavanirktok River populations (Ivishak, Lupine, Ribdon) or the Colville River population (Anaktuvuk). In the July collection, Kavik and Hula Hula site #1 fish also had positive estimates of contribution to the mixed stock sample. For the July sampling period, the sample size was smaller, and the confidence intervals were wider. Though the error terms are large, the presence of additional stocks in the July collection is realistic given what is known about offshore char migration in the Beaufort Sea area in summer.

Arctic char migrate from freshwater rivers to offshore feeding areas in early summer. There they mix in feeding aggregations, and

DRAFT

11/29/88

return to spawn and/or overwinter in late summer. Because of the small size of captured char, it is not surprising that fish caught in large enough concentrations for GSI nearshore are apparently juveniles from the Sagavanirktok River system. High apparent contributions from the Colville system are interesting. Simulations show that Anaktuvuk char are effectively identified using GSI methods, so the presence of Anaktuvuk char near Prudhoe Bay should be considered.

57

Populations with larger percentage estimated contributions to the mixed stock have relatively smaller standard errors, emphasizing the fact that the program works best with a few major contributors. With a 95% confidence interval, only Ivishak and Lupine fish persist in the June and August mixtures.

Because of the large errors associated with small estimates, it is not possible to determine conclusively whether percent composition estimates from the other populations are real, e.g., the presence of Babbage char in Prudhoe Bay in the July sample. Only Lupine and Anaktuvuk River char estimates are non-zero in a 95% confidence interval in this collection.

From simulations we know that Canning River char are underestimated, and are probably assigned to the Ivishak River population which it resembles in genetic characteristics. It is possible that those identified as Ivishak fish in the Endicott collections are actually Canning char.

Through predominance of Sagavanirktok and Anaktuvuk fish in non-zero estimates is reasonable considering what is known of the biology of North Slope Arctic char, the estimates would be more useful

if the standard errors around the means of the estimates were smaller. The mixed stock sample sizes obtained are probably all too small for a 14-stock baseline. The work by Wood et al. (1987) shows with simulations that a mixed stock collection should contain 50 samples for each baseline. For computational ease, his work only included three stocks. Other researchers, such as Milner et al. (198?), using more than 20 stocks, have found that this rule is too extreme. The sample sizes necessary to answer specific questions can be calculated from the empirically-determined level of divergence in the target species, the number of variable loci, and the management goals for precision. The relationship of number of samples needed is not an arithmatic function of the number of baselines, but rather a function of the level of divergence of the species, and what is acceptable to management.

58

The basis of genetic stock identification is electrophoretically detectable differences in genotype frequencies between stocks. To do genetic stock identification (GSI) there must be sufficient detectable genetic variation in the stocks to be studied. Variation between groups of populations, e.g., between those of major drainages, should be relatively high combined with a low within-group variability. Also, the baseline should represent the major populations contributing to the mixed stock to be analyzed.

The level of divergence among populations that is detected using electrophoretic methods depends on the species and area. Different species have different levels of detectable differences (e.g., sockeye salmon are low, chinook are typically high). The species in question

may be at the center or edge of its range, and the relationships among populations of a species may reflect the evolutionary history of that species is in that area. Isolated populations and those colonized by a limited number or those experiencing stochastic fluctuations in number are more likely to have a low level of variability. Because of genetic drift, they may be different because of low variability or different because uncommon genotypes increased in a small population by chance.

59

The number of genetic loci that are studied can affect the accuracy and precision of genetic stock identification. Although the loci used in any study are meant to represent a random sample of the genome, and any sample of genetic characters should give similar estimates of the relationships among populations, the possibility exists that additional variable loci may introduce unique genotype combinations identifiable using maximum likelihood statistics.

Milner et al. (1983) used electrophoretic data from chinook salmon to simulate the addition of loci. They observed a 60% increase in accuracy with an increase from 10 to 25 loci. Wilmot (1988) found that the point estimates for chum salmon allocations to United States versus Canada stocks changed markedly when the number of variable loci was increased from 7 to 12.

The number of loci and the sample size needed are correlated in an inverse relationship. This relationship can be used either to increase accuracy and precision by increasing both number of characters measured and the sample size, or decrease the number of samples required to get similar levels of accuracy and precision.

60

DRAFT

### Implications for Management

North Slope Arctic char have an above average amount of genetic variation compared to other salmonids. The pattern of variation shows distinctness among different populations, but the differences do not correspond to migratory versus resident life history strategies that have been postulated.

The pattern of divergence indicated by comparisons, pairwise, by log likelihood ratio statistics (G-tests) and by a calculation of gene diversity ( $G_{ST}$ ) indicated that char populations are genetically distinct. These statistics measure diversity, but may not be good indicators of the level of divergence among populations that is required to do genetic stock identification.

Genetic stock identification techniques have potential for North Slope Arctic char biology and management, but need improvements. The reliability of the estimates is hard to verify. With little data on population dynamics from other sources, we have relied mainly on simulations for indications of the accuracy and precision in our estimates.

Simulations show that certain populations of Arctic char are allocated correctly, but that others are not. Though lack of precision is typical in composition estimates where many stocks contribute a small amount to a mixture, certain population of North Slope char are still not identified accurately in the 40 to 80% composition range. It suggests that the detected level of divergence in North Slope char populations is not sufficient to distinguish all 61

DRAFT

populations using our current baseline. This is also supported by the high levels of genetic similarity (Nei 1972) observed among certain population pairs.

Despite inaccuracies and imprecision we have observed in our simulations with GSI for North Slope Arctic char, the allocations made by the program with actual data from mixed fishery stocks from the Endicott area are supported by biological data. The collections in June, July, and August 1987 were made near the mouth of the Sagavanirktok River. The stocks identified in these mixtures are predominatly from the Sagavanirktok River Drainage, particularly in June and August when these fish would first be outmigrating to feed, then returning to overwinter. The July sample apparently included fish from other drainages, supporting data that shows that Arctic char migrate considerable distances and mix offshore during the summer season.

It is possible to improve both the accuracy and precision of GSI estimates. This can be done by increasing the completeness of the baseline; the number of loci in baseline and mixed stock samples; and the sample size of baseline and mixed fishery samples.

We believe we have major stocks, though more questions could be addressed if more collections were made within certain drainages with numerous spawning stocks, and with additional non-migratory stocks. Data from more baseline, spawning populations may need to be collected in the future to allow us to increase the number of loci in the analyses.

More loci studied would correspond to more data from each fish

DRAFT

sampled, and therefore smaller sampled sizes would be necessary to get the same level of precision. Additional loci are possible with an increase in sample quality and effort.

Increased sample sizes in mixed fishery samples improve both the accuracy and precision of GSI estimates. Increasing both the number of loci and the sample sizes would do the most for increasing both accuracy and precision in mixed stock identification procedures.

It has been determined that the Beaufort Sea environment is highly changeable, and that the dynamics of fish populations is highly dependent on wind conditions from year to year. In order to understand the distribution and timing of the migratory North Slope Arctic char it will be necessary to sample numerous places offshore, and at more times during the summer season when they migrate. This would allow us to determine how these fish use the area, predict how our activities may affect them, and use consideration for them in our plans for development.

a second a second and the second and the second second and the second second second second second second second

DRAFT

#### LITERATURE CITED

- Allendorf, F. W. and S. R. Phelps. 1981. Use of allelic frequencies to describe population structure. Canadian Journal of Fisheries and Aquatic Sciences 38:1507-1514.
- Allendorf, F. W. and G. H. Thorgaard. 1984. Tetraploidy and the evolution of salmonid fishes. Pages 1-53 <u>in</u> B. J. Turner, editor. Evolutionary genetics of fishes. Plenum Press, NY.
- Allendorf, F. W., K. L. Knudsen, and R. F. Leary. 1983. Adaptive significance of differences in the tissue-specific expression of a phosphoglucomutase gene in rainbow trout. Proceedings of the National Academy of Sciences U.S.A. 80:1397-1400.
- Allendorf, F. W. and F. M. Utter. 1979. Population genetics. Pages 407-454 in W. S. Hoar, S. S. Randall and J. R. Brett, editors. Fish physiology. Academic Press, NY.
- Allendorf, F. W., N. Mitchell, N. Ryman, and G. Stahl. 1977. Isozyme loci in brown trout (<u>Salmo trutta</u>): detection and interpretation from population data. Hereditas 86:179-190.
- Andersson, L., N. Ryman, and G. Stahl. 1983. Protein loci in the Arctic charr, <u>Salvelinus alpinus</u> L.: electrophoretic expression and genetic variability patterns. Journal of Fish Biology 23:75-94.
- Ayala, F. J. and J. A. Kiger, Jr. 1980. Modern genetics. The Benjamin/Cummings Publishing Company, Inc. Menlo Prk, CA.
- Bain, L. H. 1974. Life histories of three species of freshwater fishes in Beaufort Sea Drainages, Yukon Territory. P. J. McCart, editor. Aquatic Environments Limited. Canadian Arctic Gas Study Limited/Alaskan Arctic Gas Study Company Biological Report Series Volume Eighteen.
- Beacham, T. D., R. E. Withler, and A. P. Gould. 1985. Biochemical genetic stock identification of chum salmon (<u>Oncorhynchus keta</u>) in southern British Columbia. Canadian Journal of Fisheries and Aquatic Sciences 42:437-448.
- Boyer, S. H., D. C. Fainer, and M. A. Naughton. 1963. Myoglobin: inherited structural variation in man. Science 140:1228-1231.
- Chakraborty, R. 1980. Gene diversity analysis in nested subdivided populations. Genetics 96:721-726.
- Clayton, J. W. and D. N. Tretiak. 1972. Amine citrate buffers for pH control in starch gel electrophoresis. Journal of the Fisheries Research Board of Canada 29:1169-1172.

Cooper, D. W. 1968. The significance level in multiple tests made simultaneously. Heredity 23:614-617.

- Craig, P. C. 1977. Fisheries investigations along the North Slope and Beaufort Sea coast in Alaska with emphasis on Arctic char. P. J. McCart, editor. Aquatic Environments Limited. Canadian Arctic Gas Study Limited/Alaskan Arctic Gas Study Company Biological Report Series Volume Forty-one.
- Craig, P. C. and P. J. McCart. 1975. Fish utilization of nearshore coastal waters between the Colville and Mackenzie Rivers with an emphasis on anadromous species. Pages 172-219 in P. C. Craig, editor. Fisheries investigations in a coastal region of the Beaufort Sea. Canadian Arctic Gas Study Limited/Alaskan Arctic Gas Study Company Biological Report Series 34.
- Craig, P. C. and P. McCart. 1976. Fish use of nearshore coastal waters in the western arctic: emphasis on anadromous species. Pages 361-388 in D. W. Hood and D. C. Burnell, editors. Assessment of the Arctic marine environment: Selected topics. University of Alaska, Fairbanks, Alaska. Institute of Marine Sciences Occasional Publication 4.
- Everett, R. J. and R. L. Wilmot. 1987. Population genetic structure of Arctic char (<u>Salvelinus alpinus</u>) from rivers of the North Slope of Alaska. U.S. Fish and Wildlife Service, Anchorage, AK. Arctic Fish Habitats and Sensitivities Study, funded by U.S. Minerals Management Service through National Oceanic and Atmospheric Administration.
- Furniss, R. A. 1975. Inventory and cataloging of Arctic area waters. Annual report of progress, 1974-1975. Federal Aid in Fish Restoration. Sport Fish Investigations of Alaska. Alaska Department of Fish and Game. Project F-9-6, Study G-I.
- Glova, G. and P. J. McCart. 1974. Life history of Arctic char (<u>Salvelinus alpinus</u>) in the Firth River, Yukon Territory. Pages 1-50 <u>in</u> P. J. McCart, editor. Life Histories of Anadromous and Freshwater Fish in the Western Arctic. Canadian Arctic Gas Study Limited/Alaskan Arctic Gas Study Company Biological Report Series 20.
- Hartl, D. 1980. Principles of population genetics. Sinauer Associates, Sunderland, MA.
- Johnson, K. R. 1984. Protein variation in Salmoninae: genetic interpretations of electrophoretic banding patterns, linkage associations among loci, and evolutionary relationships among species. Ph.D. Thesis, The Pennsylvania State University.

- Jonsson, B. and K. Hindar. 1982. Reproductive strategy of dwarf and normal Arctic charr (<u>Salvelinus alpinus</u>) from Vangsvatnet Lake, western Norway. Canadian Journal of Fisheries and Aquatic Sciences 39:1404-1413.
- McCart, P. and P. Craig. 1971. Meristic differences between anadromous and freshwater-resident Arctic char (<u>Salvelinus</u> <u>alpinus</u>) in the Sagavanirktok River drainage, Alaska. Journal of the Fisheries Research Board of Canada 28:115-118.
- Millar, R. B. 1987. Maximum likelihood estimation of mixed stock fishery composition. Canadian Journal of Fisheries and Aquatic Sciences 44:583-590.
- Milner, G. B., D. J. Teel, P. B. Aebersold, and F. M. Utter. 1986. Genetic stock identification. Annual report of research (Fiscal Year 1985), National Ocean and Atmospheric Administration, Northwest and Alaska Fisheries Center, Seattle, WA.
- Milner, G. B., D. J. Teel, F. M. Utter, and C. L. Burley. 1981. Columbia River stock identification study: Validation of genetic method. Annual report of research (Fiscal Year 1980), National Ocean and Atmospheric Administration, Northwest and Alaska Fisheries Center, Seattle, WA.
- Nei, M. 1972. Genetic distance between populations. American Naturalist 196:283-292.
- Nei, M. 1973. Analysis of gene diversity in subdivided populations. Proceedings of the National Academy of Sciences U.S.A. 70:3321-3323.
- Nei, M. 1978. Estimation of average heterozygosity and genetic distance from a small number of individuals. Genetics 89:583-590.
- Nevo, E. 1978. Genetic variation in natural populations: patterns and theory. Theor. Pop. Biol. 13:121-177.
- Nordeng. H. 1983. Solution to the "char problem" based on Arctic char (<u>Salvelinus alpinus</u>) in Norway. Canadian Journal of Fisheries and Aquatic Sciences 40:1372-1387.
- Ridgway, G. J., S. W. Sherburne, and R. D. Lewis. 1970. Polymorphisms in the esterases of Atlantic herring. Transactions of the American Fisheries Society 99:147-151.
- Ryman, N. and G. Stahl. 1981. Genetic perspectives of the identification and conservations of Scandinavian stocks of fish. Canadian Journal of Fisheries and Aquatic Sciences 38:1562-1575.
- Sneath, P. H. A. and R. R. Sokal. 1973. Numerical Taxonomy. W. H. Freeman, San Francisco.
- Sokal, R. R. and F. J. Rohlf. 1981. Biometry (2nd edition). W. H. Freeman, San Francisco, CA.
- Utter, F. M., H. O. Hodgins, and F. W. Allendorf. 1974. Biochemical genetic studies of fishes: potentialities and limitations. Pages 213-238 <u>in</u> D. C. Malins and J. R. Sargent, editors. Biochemical and biophysical perspectives in marine biology. Volume 1. Academic Press, NY.
- Utter, F. M., D. Campton, S. Grant, G. Milner, J. Seeb, and L. Wishard. 1981. Population structures of indigenous salmonid species of the Pacific Northwest. Pages 285-304 <u>in</u> W. J. McNeil and D. C. Hinsworth, editors. Salmonid ecosystems of the North Pacific. Oregon State University Press, Corvallis, Oregon.
- Wood, C. D., S. McKinnell, T. J. Mulligan, and D. A. Fournier. 1987. Stock identification with the maximum-likelihood mixture model: sensitivity analysis and application to complex problems. Canadian Journal of Fisheries and Aquatic Sciences 44:866-881.

Appendix A.- Gene frequencies of variable loci in 12 populations of Arctic char collected in 1987 from the North Slope of Alaska and Canada. Variants of duplicated loci were arbitrarily assigned to one locus of the duplicated pair. Names of enzyme loci (abbreviated here) are in Table 2.

					in the		Popula	ations					
Loci		AICH	BAB1	BAB2	CAN1	CAN2	FIR1	FIR2	HULA	KONG	LUPI	SADL	SHUB
AAT1	100 33 N	0.975 0.025 40.00	1.000 0.000 53.00	1.000 0.000 21.00	0.975 0.025 40.00	0.956 0.044 45.00	ND 	0.947 0.053 38.00	0.981 0.019 80.00	1.000 0.000 45.00	1.000 0.000 45.00	1.000 0.000 44.00	0.956
AAT3	100 75 129 N	0.911 0.089 0.000 45.00	1.000 0.000 0.000 53.00	1.000 0.000 0.000 21.00	0.910 0.090 0.000 50.00	0.936 0.064 0.000 55.00	0.920 0.080 0.000 44.00	0.956 0.033 0.011 45.00	0.950 0.050 0.000 80.00	0.932 0.068 0.000 44.00	0.956 0.044 0.000 45.00	0.867 0.133 0.000 45.00	0.922 0.078 0.000 45.00
ACO4	100 115 130 N	0.533 0.211 0.256 45.00	0.490 0.019 0.490 52.00	0.441 0.000 0.559 17.00	0.477 0.244 0.279 43.00	0.611 0.167 0.222 45.00	0.434 0.196 0.370 46.00	0.544 0.200 0.256 45.00	0.562 0.219 0.219 80.00	0.544 0.189 0.267 45.00	0.467 0.211 0.322 45.00	0.100 0.011 0.889 45.00	0.444 0.233 0.322 45.00
FH	100 130 N	0.433 0.567 45.00	ND 	ND  21.00	ND 	ND 	ND 	ND  46.00	ND 	0.422 0.578 45.00	0.619 0.381 42.00	0.444 0.556 45.00	0.578
GAP 3	100 Null N	0.716 0.284 44.00	0.933 0.067 52.00	0.850 0.150 20.00	0.744 0.256 43.00	0.756 0.244 45.00	0.767 0.233 43.00	0.826 0.174 46.00	0.581 0.419 80.00	0.682 0.318 44.00	0.767 0.233 45.00	1.000 0.000 45.00	0.738 0.262 42.00
GPI1	100 55 N	1.000 0.000 45.00	0.990 0.010 52.00	1.000 0.000 21.00	1.000 0.000 55.00	1.000 0.000 55.00	1.000 0.000 44.00	0.989 0.011 45.00	1.000 0.000 80.00	1.000 0.000 44.00	1.000 0.000 45.00	1.000 0.000 45.00	1.000 0.000 45.00
GPI3	100 96 N	0.744 0.256 45.00	0.846 0.154 52.00	1.000 0.000 21.00	0.891 0.109 55.00	0.927 0.073 55.00	0.659 0.341 44.00	0.667 0.333 45.00	0.819 0.181 80.00	0.733 0.267 45.00	0.589 0.411 45.00	0.844 0.156 45.00	0.822 0.178 45.00
IDH2	100 220 N	0.967 0.033 45.00	1.000 0.000 53.00	1.000 0.000 21.00	1.000 0.000 41.00	1.000 0.000 43.00	0.997 0.023 43.00	1.000 0.000 46.00	1.000 0.000 80.00	0.977 0.023 44.00	0.978 0.022 45.00	1.000 0.000 45.00	1.000 0.000 45.00
IDH3	100 80 N	0.889 0.111 45.00	1.00 0.000 53.00	1.00 0.000 21.00	0.977 0.023 44.00	0.978 0.022 45.00	0.935 0.065 46.00	0.946 0.054 46.00	0.975 0.025 79.00	1.000 0.000 45.00	1.000 0.000 45.00	1.000 0.000 45.00	0.944 0.056 45.00
LDH5	100 97 N	0.944 0.056 45.00	1.000 0.000 48.00	1.000 0.000 21.00	1.000 0.000 45.00	1.000 0.000 45.00	0.978 0.022 46.00	0.978 0.022 46.00	0.988 0.012 80.00	1.000 0.000 45.00	1.000 0.000 45.00	1.000 0.000 45.00	1.000 0.000 45.00
MDH1	100 128 N	1.000 0.000 45.00	1.000 0.000 53.00	1.000 0.000 21.00	1.000 0.000 43.00	1.000 0.000 45.00	1.000 0.000 41.00	1.000 0.000 46.00	1.000 0.000 80.00	1.000 0.000 45.00	1.000 0.000 40.00	1.000 0.000 45.00	1.000 0.000 45.00
ME3	100 69 N	1.000 0.000 45.00	1.000 0.000 53.00	1.000 0.000 21.00	1.000 0.000 45.00	1.000 0.000 45.00	1.000 0.000 46.00	1.000 0.000 46.00	1.000 0.000 80.00	1.000 0.000 45.00	1.000 0.000 45.00	1.000 0.000 45.00	1.000
6PG1	100 95 N	1.000 0.000 45.00	1.000 0.000 53.00	1.000 0.000 21.00	1.000 0.000 45.00	1.000 0.000 45.00	1.000 0.000 46.00	1.000 0.000 46.00	1.000 0.000 80.00	1.000 0.000 45.00	0.989 0.011 45.00	1.000 0.000 45.00	1.000
PGM2	100 88 N	1.000 0.000 45.00	1.000 0.000 51.00	1.000 0.000 21.00	1.000	1.000	1.000	1.000 0.000 44.00	0.975 0.025 80.00	1.000	1.000	1.000	1.000
SDH1	100 43 N	0.922 0.078	1.000	1.000	0.932	0.966	0.978	0.978	0.913	0.989	0.989	1.000	0.878 0.122
SOD1	100 115 87 N	0.944 0.056 0.000 45.00	0.944 0.000 0.056 53.00	0.857 0.000 0.143 21.00	0.973 0.027 0.000 55.00	0.973 0.027 0.000 50.00	1.000 0.000 0.000 46.00	1.000 0.000 0.000	0.900 0.100 0.000	0.978 0.022 0.000	0.911 0.089 0.000	1.000 0.000 0.000	45.00 0.967 0.033 0.000
XDH1	100 86 N	0.433 0.567 45.00	ND 53.00	ND	ND	ND	ND 45.00	ND 46.00	ND 	0.422 0.578 45.00	0.619 0.381 42.00	45.00 0.444 0.556 45.00	45.00 0.578 0.422 45.00
AICH = Aichilik BAB1 = Babbage Site 1 BAB2 = Rebbage Site 2				CAN1 = Canning Site 1 $CAN2 = Canning Site 2$ $FIR = First Site 2$			HULA = Hula Hula KONG = Kongakut			SADL = Sadlerochit SHUB = Shublik			

Appendix B.- Gene frequencies of variable loci in 16 populations of Arctic char collected in 1986 and 1987 from the North Slope of Alaska and Canada. Variants of duplicated loci were arbitrarily assigned to one locus of the duplicated pair. Names of enzyme loci (abbreviated here) are in Table 2.

	Populations															
	AIC	ANA	BA1	BA2	CAN	CAO	EGA	FIR	HU1	HU2	IVI	KAV	KON	LUP	RIB	SAD
100 33 N	0.988 0.012 80	0.975 0.025 40	1.000 0.000 53	1.000 0.000 33	0.969 0.031 179	1.000 0.000 21	0.986 0.014 35	0.921 0.079 76	0.981 0.019 80	ND 	1.000 0.000 22	1.000 0.000 37	1.000 0.000 85	1.000 0.000 45	ND 	1.000 0.000 44
100 75 129 N	0.918 0.082 0.000 85	0.908 0.092 0.000 38	1.000 0.000 0.000 53	1.000 0.000 0.000 35	0.908 0.092 0.000 206	1.000 0.000 0.000 21	0.951 0.049 0.000 41	0.942 0.050 0.008 130	0.950 0.050 0.000 80	ND  00	0.966 0.034 0.000 72	ND  00	0.912 0.088 0.000 85	0.956 0.044 0.000 45	0.950 0.050 0.000 40	0.867 0.133 0.000 45
100	0.541	0.403	0.490	0.700	0.514	0.441	0.557	0.461	0.560	0.467	0.535	0.463	0.547	0.467	0.588	0.100
115	0.194	0.292	0.019	0.029	0.216	0.000	0.243	0.211	0.220	0.239	0.174	0.137	0.177	0.211	0.112	0.011
130	0.265	0.305	0.490	0.271	0.270	0.559	0.200	0.328	0.220	0.294	0.291	0.400	0.275	0.322	0.300	0.889
N	85	36	52	35	183	17	35	128	91	46	72	40	85	45	40	45
100 Null N	0.655 0.345 82	0.934 0.066 38	0.933 0.067 52	ND 	0.754 0.246 183	0.850 0.150 20	0.500 0.500 32	0.883 0.177 107	0.560 0.440 91	0.325 0.675 40	0.775 0.225 71	0.706 0.294 34	0.714 0.286 84	0.767 0.233 45	0.730 0.270 37	1.000 0.000 45
100	1.000	0.950	0.990	1.000	0.998	1.000	1.000	0.981	1.000	1.000	0.993	1.000	1.000	1.000	0.923	1.000
55	0.000	0.050	0.010	0.000	0.002	0.000	0.000	0.019	0.000	0.000	0.007	0.000	0.000	0.000	0.077	0.000
N	85	40	52	35	211	21	41	129	95	51	74	40	85	45	39	45
100	0.759	0.900	0.846	0.629	0.897	1.000	0.829	0.694	0.842	0.860	0.912	0.988	0.718	0.589	0.667	0.844
96	0.241	0.100	0.154	0.371	0.123	0.000	0.171	0.306	0.158	0.140	0.088	0.012	0.282	0.411	0.333	0.156
N	85	39	52	35	211	21	41	129	95	50	74	40	85	45	39	45
100	0.982	1.000	1.000	1.000	0.997	1.000	1.000	0.981	1.000	0.990	0.986	0.975	0.988	0.978	0.950	1.000
220	0.018	0.000	0.000	0.000	0.003	0.000	0.000	0.019	0.000	0.010	0.014	0.025	0.012	0.022	0.050	0.000
N	85	37	53	35	149	21	35	129	95	51	74	40	85	45	40	45
100	0.888	1.000	1.000	1.000	0.965	1.000	0.986	0.947	0.973	0.927	0.993	0.900	0.941	1.000	1.000	1.000
80	0.122	0.000	0.000	0.000	0.035	0.000	0.014	0.053	0.027	0.073	0.007	0.100	0.059	0.000	0.000	0.000
N	85	39	53	35	184	21	35	131	91	48	73	40	85	45	40	45
100	0.946	1.000	1.000	0.986	0.995	1.000	0.929	0.973	0.989	0.941	0.973	0.988	0.965	1.000	0.923	1.000
97	0.054	0.000	0.000	0.014	0.005	0.000	0.071	0.027	0.011	0.059	0.027	0.012	0.035	0.000	0.077	0.000
N	83	35	53	35	188	21	35	132	95	51	73	40	85	45	39	45
100	1.000	0.956	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000
128	0.000	0.044	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
N	82	34	53	35	182	21	35	128	93	52	72	40	85	40	40	45
100	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	0.981	1.000	1.000	1.000	1.000	1.000	1.000
69	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.019	0.000	0.000	0.000	0.000	0.000	0.000
N	85	40	53	35	197	21	35	132	95	54	74	40	85	45	40	45
100	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	0.993	1.000	1.000	0.989	0.988	1.000
95	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.007	0.000	0.000	0.011	0.012	0.000
N	85	40	53	35	197	21	35	132	95	51	74	40	85	45	40	45
100	1.000	1.000	1.000	1.000	1.000	1.000	1.000	0.996	0.979	0.971	1.000	1.000	1.000	1.000	1.000	1.000
88	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.004	0.021	0.029	0.000	0.000	0.000	0.000	0.000	0.000
N	85	35	51	28	161	21	39	128	95	51	50	40	85	45	40	45
100	0.965	0.875	1.000	1.000	0.912	1.000	0.986	0.985	0.914	1.000	0.913	1.000	0.959	0.989	1.000	1.000
43	0.035	0.125	0.000	0.000	0.088	0.000	0.014	0.015	0.086	0.000	0.087	0.000	0.041	0.011	0.000	0.000
N	85	36	53	34	188	21	35	131	87	17	23	40	85	44	40	44
100 115 87 N	0.947 0.053 0.000 85	1.000 0.000 0.000 35	0.944	0.957 0.015 0.028 35	0.974 0.026 0.000 212	0.857 0.000 0.143 21	0.943	0.996 0.004 0.000 132	0.914 0.086 0.000 93	0.907 0.093 0.000 54	0.972 0.028 0.000 72	1.000 0.000 0.000 40	0.976 0.024 0.000 85	0.911 0.089 0.000 45	0.888 0.112 0.000 40	1.000 0.000 45
	100 33 N 100 75 129 N 100 115 130 N 100 55 N 100 220 N 100 220 N 100 220 N 100 220 N 100 220 N 100 220 N 100 220 N 100 220 N 100 220 N 100 55 N 100 96 N 100 200 N 100 100 N 100 100 N 100 100 N N 100 N N N 100 N N N N	AIC           100         0.988           33         0.012           N         80           100         0.918           75         0.082           129         0.000           N         85           100         0.541           115         0.194           130         0.265           N         82           100         0.655           Null         0.345           N         82           100         1.000           55         0.000           N         85           100         0.759           96         0.241           N         85           100         0.982           220         0.018           N         85           100         0.946           97         0.054           N         83           100         1.000           1.000         946           97         0.054           N         85           100         1.000           1.000         9.0000           N	AIC         ANA           100         0.988         0.975           33         0.012         0.025           N         80         40           100         0.918         0.908           75         0.082         0.092           129         0.000         0.000           N         85         38           100         0.541         0.403           115         0.194         0.292           130         0.265         0.305           N         85         36           100         0.655         0.934           Null         0.345         0.066           N         82         38           100         1.655         0.934           Null         0.345         0.066           N         82         38           100         1.000         0.950           55         0.000         0.050           N         85         39           100         0.982         1.000           N         85         37           100         0.988         1.000           N         85         39	AIC         ANA         BA1           100         0.988         0.975         1.000           33         0.012         0.025         0.000           N         80         40         53           100         0.918         0.908         1.000           75         0.082         0.092         0.000           129         0.000         0.000         0.000           129         0.000         0.000         0.000           N         85         38         53           100         0.541         0.403         0.490           115         0.194         0.292         0.019           130         0.265         0.305         0.490           N         85         36         52           100         0.655         0.934         0.933           Null         0.345         0.066         0.067           N         82         38         52           100         1.000         0.950         0.990           55         0.000         0.050         0.010           N         85         39         52           100         0.982	AIC         ANA         BA1         BA2           100         0.988         0.975         1.000         1.000           33         0.012         0.025         0.000         0.000           N         80         40         53         33           100         0.918         0.902         0.000         0.000           75         0.082         0.992         0.000         0.000           129         0.000         0.000         0.000         0.000           129         0.000         0.000         0.000         0.000           15         0.194         0.292         0.019         0.029           130         0.265         0.305         0.490         0.271           N         85         36         52         35           100         0.655         0.934         0.933         ND           Null         0.345         0.066         0.067            N         82         38         52         35           100         1.000         0.950         0.990         1.000           N         85         39         52         35           10	AIC         ANA         BA1         BA2         CAN           100         0.988         0.975         1.000         1.000         0.969           33         0.012         0.025         0.000         0.000         0.031           N         80         40         53         33         179           100         0.918         0.000         1.000         0.908           129         0.000         0.000         0.000         0.000           129         0.000         0.000         0.000         0.000           100         0.541         0.403         0.490         0.271         0.216           130         0.265         0.305         0.490         0.271         0.270           N         85         36         52         35         183           100         0.655         0.934         0.933         ND         0.754           Null         0.345         0.666         0.667         -         0.246           N         82         38         52         35         211           100         1.000         0.950         0.990         1.000         0.929           220	AIC         ANA         BA1         BA2         CAN         CAO           100 $0.988$ $0.975$ $1.000$ $1.000$ $0.969$ $1.000$ 100 $0.988$ $0.975$ $1.000$ $0.000$ $0.000$ $0.000$ $0.000$ N         80         40         53         33 $179$ $21$ 100 $0.918$ $0.908$ $1.000$ $1.000$ $0.902$ $0.000$ 129 $0.000$ $0.000$ $0.000$ $0.000$ $0.000$ $0.000$ N         85         38         53         35 $206$ $21$ 100 $0.551$ $0.430$ $0.490$ $0.270$ $0.559$ N         85         36         52         35 $183$ $17$ 100 $0.655$ $0.934$ $0.933$ ND $0.754$ $0.850$ Null $0.345$ $0.666$ $0.67$ $-0.246$ $0.150$ N         82         38         52         35	AIC         ANA         BA1         BA2         CAN         CAO         EGA           100         0.988         0.975         1.000         1.000         0.969         1.000         0.986           33         0.012         0.025         0.000         0.000         0.031         0.000         0.014           N         80         40         53         33         179         21         35           100         0.918         0.902         0.000         0.231         1.010         0.245         0.305         0.490         0.271         0.270         0.550         0.500         N         82         38         52         00         183         17         35           100         0.655         0.934         0.933         ND         0.754         0.850         0.500         N	AIC         ANA         BA1         BA2         CAN         CAO         EGA         FIR           100         0.988         0.975         1.000         1.000         0.991         1.000         0.986         0.921           33         0.012         0.025         0.000         0.0031         0.000         0.014         0.079           N         80         40         53         33         179         21         35         76           100         0.918         0.908         1.000         0.908         1.000         0.900         0.000         0.900         0.000         0.900         0.000         0.902         0.000         0.000         0.000         0.200         0.000         0.200         0.200         0.200         0.201         0.211         1.1         130           100         0.541         0.403         0.490         0.211         0.214         0.150         0.500         0.328           Nul1         0.345         0.666         0.627         0.281         0.177         N         82         38         52         35         211         21         41         129           100         1.050         0.500	AIC         ANA         BA1         BA2         CAN         CAO         EGA         FIR         HU1           100         0.988         0.975         1.000         1.000         0.969         1.000         0.986         0.921         0.981           33         0.012         0.025         0.000         0.000         0.031         0.000         0.014         0.079         0.019           N         80         40         53         33         179         21         35         76         80           100         0.918         0.908         1.000         0.000	AIC         ANA         BA1         BA2         CAN         CAO         EGA         FIR         HU1         HU2           100         0.988         0.975         1.000         1.000         0.969         1.000         0.986         0.921         0.981         ND           33         0.012         0.025         0.000         0.000         0.001         0.001         0.001         0.001         0.001         0.001         0.001         0.001         0.001         0.001         0.000         0.021         0.220         0.220         0.220         0.220         0.220         0.220         0.220         0.220         0.220         0.239         0.241         0.350         0.500         0.231         0.200         0.231         0.200         0.231         0.210	AIC         ANA         BA1         BA2         CAN         CAO         EGA         FIR         HU1         HU2         IVI           100         0.988         0.975         1.000         1.000         0.986         0.921         0.981         ND         1.000           31         0.012         0.000         0.000         0.001         0.000	AIC         ANA         BA1         BA2         CAN         CAO         EGA         FIR         HU1         HU2         IVI         KAV           100         0.988         0.975         1.000         1.000         0.969         1.000         0.986         0.921         0.981         ND         1.000         1.000         0.000           31         0.012         0.025         0.000         0.000         0.000         0.001         0.007         0.000         0.000         0.000         0.000         0.000         0.000         0.000         0.000         0.000         0.000         0.000         0.000         0.000         0.000         0.000         0.000         0.000         -         0.000         -         0.000         -         0.000         -         0.034         -         1         100         0.541         0.400         0.700         0.514         0.441         0.557         0.461         0.550         0.467         0.535         0.463           100         0.655         0.300         0.200         0.328         0.200         0.240         0.251         0.775         0.766           110         0.455         0.660         0.627         0.740 </td <td><math display="block"> \begin{array}{ c c c c c c c c c c c c c c c c c c c</math></td> <td><math display="block"> \begin{array}{ c c c c c c c c c c c c c c c c c c c</math></td> <td>AIC         ANA         BA1         BA2         CAN         CAO         EGA         FIR         EU1         EU2         IVI         KAV         KON         LUP         RIB           100         0.988         0.975         1.000         1.000         0.031         0.000         0.014         0.079         0.019         -         0.000</td>	$ \begin{array}{ c c c c c c c c c c c c c c c c c c c$	$ \begin{array}{ c c c c c c c c c c c c c c c c c c c$	AIC         ANA         BA1         BA2         CAN         CAO         EGA         FIR         EU1         EU2         IVI         KAV         KON         LUP         RIB           100         0.988         0.975         1.000         1.000         0.031         0.000         0.014         0.079         0.019         -         0.000

EGA= Egakarak FIR= Firth HUI= Hula Hula Site 1 HU2= B	Hula Hula Site 2 IVI= Ivishak KAV= Kavik
KON= Konzacut LUP= Lupine RIB= Ribdon SAD= S	Sadlerochit

Appendix C.- Mean estimate (with one bootstrap standard deviation) and actual proportion of an artificial mixed stock analyzed with maximum likelihood method of genetic stock identification. Estimates are listed in descending order, and those below the line are less than 1%.

	Population	Mean esti- mate	Standard devia- tion	Actual propor- tion	80% Con inte	fidence rval	95% Con inte	fidence rval
1	Firth River	0.1874	0.0359	0.128	0.1415	0.2334	0.1170	0.2578
2	Aichilik	0.1756	0.0588	0.082	0.1003	0.2508	0.0603	0.2908
3	Hula Hula 1	0.1594	0.0517	0.092	0.0932	0.2255	0.0580	0.2607
4	Ivishak	0.1462	0.0507	0.072	0.0813	0.2111	0.0468	0.2456
5	Kongakut	0.1014	0.0756	0.082	0.0046	0.1981	-0.0468	0.2495
6	Babbage	0.0518	0.0197	0.051	0.0266	0.0771	0.0132	0.0905
7	Canning	0.0442	0.0648	0.205	-0.0388	0.1271	-0.0828	0.1712
8	Egaksrak	0.0353	0.0385	0.040	-0.0139	0.0846	-0.0401	0.1108
9	Lupine	0.0352	0.0289	0.044	-0.0018	0.0722	-0.0214	0.0919
10	Kavik	0.0323	0.0249	0.039	0.0004	0.0642	-0.0165	0.0811
11	Anaktuvuk	0.0246	0.0200	0.039	-0.0010	0.0502	-0.0146	0.0638
								• • • • • • •
12	Hula Hula 2	0.0022	0.0016	0.053	0.0002	0.0043	-0.0009	0.0054
13	Ribdon	0.0022	0.0015	0.039	0.0003	0.0041	-0.0007	0.0052
14	Canoe	0.0022	0.0014	0.034	0.0004	0.0040	-0.0006	0.0049

	Site	1	2	3	4	5	6	7	8	9	10	11	12	13	14
1	Canoe R.	84.7	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
2	Babbage-87	13.5	95.8	2.2	. 2.2	0.0	0.0	0.3	0.0	0.0	2.7	0.2	0.1	3.8	0.0
3	Firth	0.0	0.8	91.5	0.0	0.0	2.0	0.1	0.0	3.4	0.0	0.0	0.0	0.0	0.2
4	Kongakut	0.0	0.0	1.7	78.4	1.7	9.5	0.4	0.4	0.5	0.0	0.0	0.1	4.1	0.0
5	Egaksrak	0.7	0.0	2.3	4.3	92.8	7.0	8.2	6.8	0.3	0.3	1.1	3.4	0.0	0.0
6	Aichilik	0.0	0.0	0.4	8.1	0.6	79.7	0.2	0.3	0.0	0.0	0.0	0.0	0.0	0.0
7	Hula-01	0.0	0.0	0.2	1.5	0.9	0.6	86.5	0.3	0.0	0.0	0.5	0.0	0.3	0.0
8	Hula-02	0.0	0.0	0.0	0.0	0.0	0.0	0.0	90.4	0.0	0.0	0.0	0.0	0.0	0.0
9	Canning	0.0	0.0	0.1	2.8	0.8	0.4	3.6	0.0	88.0	0.0	2.0	0.0	0.1	0.7
10	Kavik	0.8	1.7	0.0	4.5	1.2	0.6	0.4	1.5	2.4	96.4	1.4	0.0	4.9	0.0
11	Ivishak	0.2	0.0	0.3	1.1	0.0	0.0	0.0	0.0	3.8	0.4	92.2	0.1	1.1	0.8
12	Ribdon	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.3	84.1	0.0	0.0
13	Lupine	0.1	2.9	0.6	1.2	0.7	0.1	0.3	0.3	0.5	0.0	1.2	12.2	92.8	0.0
14	Anaktuvuk	0.0	1.2	0.7	1.9	1.0	0.0	0.0	0.0	0.6	0.2	1.2	0.0	0.1	98.3

. 1

Appendix D.- Percentage allocations to each of 14 North Slope Arctic char stocks when an artificial mixture of each stock is compared to baseline data using the maximum likelihood method of genetic stock identification.

Appendix E.- Mean estimates (with one bootstrap standard deviation) of the composition of the June sample of Arctic char (N = 208) taken from near the Endicott causeway. Estimates are listed in descending order, and those below the line are less than 1%.

	Population	Mean estimate	Standard deviation	80% Confidence interval		95% Confidenc interval		
1	Ivishak	0.3346	0.1300	0.1682	0.5010	0.080	0.589	
2	Lupine	0.3259	0.1081	0.1875	0.4643	0.114	0.538	
3	Anaktuvuk	0.1409	0.0852	0.0318	0.2500	-0.026	0.308	
4	Kongakut	0.1239	0.1158	-0.0243	0.2721	-0.103	0.351	
5	Firth	0.0321	0.0708	-0.0585	0.1227	-0.107	0.171	
6	Aichilik	0.191	0.0255	-0.0135	0.0517	-0.031	0.069	
7	Hula Hula 1	0.0169	0.0541	-0.0523	0.0861	-0.089	0.123	
• • •								
8	Canoe	0.0009	0.0023	-0.0020	0.0038	-0.003	0.005	
9	Babbage	0.0009	0.0349	-0.0438	0.0456	-0.068	0.069	
10	Egaksrak	0.0009	0.0539	-0.0681	0.0699	-0.105	0.107	
11	Hula Hula 2	0.0009	0.0023	-0.0020	0.0038	-0.004	0.005	
12	Canning	0.0009	0.0842	-0.1069	0.1087	-0.164	0.166	
13	Kavik	0.0009	0.0175	-0.0215	0.0233	-0.033	0.035	
14	Ribdon	0.0009	0.0023	-0.0020	0.0038	-0.004	0.005	

11/29/88

DRAFT

Appendix F.- Mean estimates (with one bootstrap standard deviation) of the composition of the July sample of Arctic char (N = 126) taken from near the Endicott causeway. Estimates are listed in descending order, and those below the line are less than 1%.

	Population	Mean estimate	Standard deviation	80% Confidence interval		95% Con inte	fidence rval
144 144 144							
1	Lupine	0.2993	0.1417	0.1180	0.4807	0.0216	0.5771
2	Anaktuvuk	0.2103	0.0663	0.1255	0.2952	0.0804	0.3403
3	Hula Hula 1	0.1236	0.0798	0.0215	0.2258	-0.0328	0.2800
4	Kavik	0.1029	0.0650	0.0197	0.1861	-0.0245	0.2303
5	Babbage	0.0917	0.0786	-0.0089	0.1923	-0.0623	0.2458
6	Ribdon	0.0842	0.0786	-0.0164	0.1848	-0.0698	0.2383
7	Ivishak	0.0412	0.1591	-0.1624	0.2449	-0.2706	0.3530
•••		•••••	• • • • • • • • • • • •	•••••	• • • • • • • •	••••	•••••
8	Canoe	0.0067	0.0046	0.0008	0.0125	-0.0024	0.0157
9	Firth	0.0067	0.1088	-0.1326	0.1459	-0.2066	0.2199
10	Kongakut	0.0067	0.0315	-0.0337	0.0470	-0.0551	0.0684
11	Egaksrak	0.0067	0.0201	-0.0191	0.0324	-0.0327	0.0461
12	Aichilik	0.0067	0.0387	-0.0429	0.0562	-0.0692	0.0825
13	Hula Hula 2	0.0067	0.0046	0.0008	0.0125	-0.0024	0.0157
14	Canning	0.0067	0.1335	-0.1642	0.1775	-0.2550	0.2683

72

The head was a second to be the second to be a second to be a

DRAFT

Appendix G.- Mean estimates (with one bootstrap standard deviation) of the composition of the August sample of Arctic char (N = 126) taken from near the Endicott causeway. Estimates are listed in descending order, and those below the line are less than 1%.

73

	Population	Mean estimate	Standard deviation	80% Confidence interval		95% Con inte	fidence rval
1	Ivishak	0.4345	0.1180	0.2835	0.5856	0.2033	0.6658
2	Lupine	0.2355	0.1110	0.0934	0.3775	0.0179	0.4530
3	Ribdon	0.1075	0.0790	0.0063	0.2086	-0.0474	0.2623
4	Hula Hula 1	0.0761	0.0801	-0.0264	0.1768	-0.0809	0.2331
5	Firth	0.0650	0.0762	-0.0325	0.1625	-0.0843	0.2144
6	Egaksrak	0.0509	0.0528	-0.0166	0.1185	-0.0525	0.1544
7	Anaktuvuk	0.0223	0.0292	-0.0151	0.0597	-0.0350	0.0795
• • •		•••••					
8	Babbage	0.0012	0.0210	-0.0257	0.0281	-0.0400	0.0423
9	Canoe	0.0012	0.0058	-0.0063	0.0086	-0.0102	0.0125
10	Kongakut	0.0012	0.0327	-0.0407	0.0430	-0.0629	0.0653
11	Aichilik	0.0012	0.0059	-0.0064	0.0087	-0.0104	0.0127
12	Hula Hula 2	0.0012	0.0058	-0.0063	0.0086	-0.0102	0.0125
13	Canning	0.0012	0.0315	-0.0392	0.0415	-0.0606	0.0629
14	Kavik	0.0012	0.0211	-0.0258	0.0282	-0.0402	0.0425

## U.S. DEPARTMENT OF THE INTERIOR FISH AND WILDLIFE SERVICE

REPLY TO:

U.S. Fish and Wildlife Service Alaska Field Station-NFRC 1011 East Tudor Road Anchorage, Alaska 99503

**OFFICIAL BUSINESS** 

PENALTY FOR PRIVATE USE, \$300

\*

POSTAGE AND FEES PAID U.S. DEPARTMENT OF THE INTERIOR INT-423



AK electrephoneris

Thing vellowith chom CJ 7 AS 45(9)

DR. ROBERT BEHNKE DEPARTMENT OF FISHERY & WILDLIFE BIOLOGY COLORADO STATE UNIVERSITY FORT COLLINS, COLORADO 80523

. ...