

Stock Identification of Yukon River Chinook and Chum Salmon using Microsatellite DNA

Loci

Report to Yukon River Panel : Project CRE 79-06

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Table of Contents

Abstract.....	2
Introduction.....	3
Materials and Methods.....	4
Results and Discussion.....	7
Acknowledgments.....	8
Literature Cited.....	8

List of Tables

Table 1. Baseline used to estimate stock compositions of chum and Chinook salmon from the fish wheel tagging program at Bio Island 2006.	12
Table 2. Estimated percentage stock composition of chum salmon migrating past the fish wheel tagging program at Bio Island, 2006. Stock compositions were estimated using 14 microsatellite loci and the baseline outlined in Table 1. Standard deviations of the estimate are in parentheses.	13
Table 3. Estimated percentage stock composition of Chinook salmon migrating past the fish wheel tagging program at Bio Island, 2006. Stock compositions were estimated using 13 microsatellite loci and the baseline outlined in Table 1. Standard deviations of the estimate are in parentheses.	14

Abstract

Stock identification of chum and Chinook salmon migrating past the DFO fish wheel program at Bio Island, near the Yukon-Alaska border, was conducted in 2006 through analysis of microsatellite variation. Variation at 14 microsatellite loci was surveyed for 728 chum salmon and 747 Chinook salmon collected from the fish wheel program. The seasonal sample for each species was structured so that migrating salmon were sampled in proportion to run abundance on a weekly basis.

For chum salmon, spawning populations from the White River drainage were estimated to comprise 55% of the fish migrating past the Bio Island fish wheels, while 41% were estimated to have been from mainstem Yukon River chum salmon spawning populations.

For Chinook salmon, the major regional stocks contributing to the run were Carmacks area tributaries (Big Salmon River, Little Salmon River, Tatchun Creek) (33%), Teslin River (13%), Stewart River (13%), Pelly River (12%), north Yukon mainstem tributaries (Chandindu River, Klondike River) (10%), mid-mainstem Yukon and Nordenskiold River (10%), and upper Yukon tributaries (6%).

Introduction

Chum salmon (*Oncorhynchus keta*) and Chinook salmon (*O. tshawytscha*) are widely distributed throughout the Yukon River drainage, spawning in tributaries ranging from the extreme headwaters (e.g. Teslin River, British Columbia) to near the mouth of the river (e.g. Andreafsky River, Alaska). Management for conservation of biodiversity within the drainage requires knowledge of genetic variation among populations as well as population-specific information from fisheries. Effective management of fisheries in major drainages like the Yukon River generally requires information on the harvest and timing of specific populations, should managers wish to change exploitation rates on specific populations for conservation purposes. For example, the Canada/U.S. Yukon River Salmon Agreement has established escapement targets and harvest sharing provisions for Canadian-origin salmon stocks. It is therefore important to develop a management system that allows managers to accurately assess the status of these stocks in fisheries throughout the drainage during the season so that management decisions can ensure that Treaty obligations are achieved. Accurate post-season run reconstructions are essential in evaluating whether management actions were consistent with meeting overall objectives and Treaty obligations. Run reconstructions are also important in monitoring the productivity of stocks and assessing the adequacy of current escapement targets and both pre-season forecasting and in-season run assessment techniques. Without this knowledge, managing to achieve Treaty obligations is difficult and severely limits the assessment of factors influencing stock productivity, which appear to have fluctuated widely in recent years.

Stock identification of chum and Chinook salmon migrating through the mainstem Yukon River is a continuing management concern. Although allozyme-based methods of stock identification have proven useful in the estimation of chum salmon stock composition in mixed-stock fisheries (Shaklee et al. 1999), and differentiation at allozyme loci occurs among Yukon

River chum salmon (Beacham et al. 1988; Wilmot et al. 1992), the level of population discrimination available in the Yukon River is not yet sufficient for population-specific applications. Variation in microsatellite loci has been applied in other species requiring discrimination among salmonid populations within watersheds (Small et al. 1998; Beacham and Wood 1999; Beacham et al. 2001), and has been shown to be useful in stock discrimination in Chinook salmon (Banks et al. 2000). Variation at microsatellite loci has been particularly useful for population-specific estimates of stock composition of Fraser River Chinook salmon (Beacham et al. 2003).

In 2006, we surveyed variation at 14 microsatellite loci for chum salmon and 13 microsatellite loci for Chinook salmon. Samples were obtained from salmon live-captured at the Bio Island fish wheels in the lower portion of the Yukon River in Canada. We used microsatellite variation to estimate stock composition in the samples collected.

Materials and Methods

Collection of DNA Samples and Laboratory Analysis

Tissue samples were collected from adult chum and Chinook salmon migrating past the Bio Island fishwheels in the lower Yukon River between July 6 and October 7, 2006. Samples were weighted according to stratified abundance estimates as determined from the mark-recapture program, with a target sample size of 750 fish to be analyzed for each species. Adipose punches were taken from sampled fish and DNA was extracted as described by Withler et al. (2000).

Once chum salmon genomic DNA was available, surveys of variation at the following 14 microsatellite loci were conducted: *Ots3* (Banks et al. 1999), *Oke3* (Buchholz et al. 2001), *Oki2* (Smith et al. 1998), *Oki100* (Miller et al. unpub), *Ots103* (Nelson and Beacham 1999),

Omm1070 (Rexroad et al. 2001), *Omy 1011* (Spies et al. 2005), *One101*, *One102*, *One104*, *One111*, and *One114* (Olsen et al. 2000), *Ssa419* (Cairney et al. 2000), and *OtsG68* (Williamson et al. 2002). Microsatellites were size fractionated in an Applied Biosystems (ABI) 3730 capillary DNA sequencer, and genotypes were scored by GeneMapper software 3.0 (Applied Biosystems, Foster City, CA) using an internal lane sizing standard.

For Chinook salmon, the following 13 microsatellite loci were surveyed for genetic variation: *Ots3* (Banks et al. 1999), *Ots208b* (Grieg et al. 2003), *OtsG474* (Williamson et al. 2002), *Ots212* (Grieg et al. 2003), *Oki100* (Miller et al., unpub), *Ots9* (Banks et al. 1999), *Ogo2* (Olsen et al. 1998), *Ogo4* (Olsen et al. 1998), *Omm1080* (Rexroad et al. 2002), *Ots201b* (OSU, unpub), *Ots211* (Grieg et al. 2003), *Ots213* (Grieg et al. 2003), and *Ssa408* (Cairney et al. 2000). Microsatellites were size fractionated in an ABI 3730 capillary DNA sequencer, and genotypes were scored by GeneMapper software 3.0 (Applied Biosystems, Foster City, CA) using an internal lane sizing standard.

In general, polymerase chain (PCR) reactions were conducted in 10 µl volumes consisting of 0.06 units of Taq polymerase, 1µl of 30ng DNA, 1.5-2.5mM MgCl₂, 1mM 10x buffer, 0.8mM dNTP's, 0.006-0.065µM of labeled forward primer (depending on the locus), 0.4µM unlabeled forward primer, 0.4µM unlabeled reverse primer, and deionized H₂O. PCR was completed on an MJResearch™ DNA Engine™ PCT-200 or a DNA Engine Tetrad™ PCT-225. The amplification profile involved one cycle of 2 min @ 92°C, 30 cycles of 15 sec @ 92°C, 15 sec @ 52-60°C (depending on the locus) and 30 sec @ 72°C, and a final extension for 10 min @ 72°C. Specific PCR conditions for a particular locus could vary from this general outline.

Baseline Populations

The baseline survey consisted of microsatellite analysis of chum salmon from 8 locations and microsatellite analysis of Chinook salmon from 21 locations within the Canadian portion of

the drainage (Table 1). All annual samples available for a specific sample location were combined to estimate population allele frequencies, as was recommended by Waples (1990).

Estimation of Stock Composition

Analysis of fishery samples was conducted with a Bayesian procedure (BAYES) as outlined by Pella and Masuda (2001). Each locus was assumed to be in Hardy-Weinberg equilibrium, and expected genotypic frequencies were determined from the observed allele frequencies and used as model inputs. For BAYES, the initial FORTRAN-based computer program as outlined by Pella and Masuda (2001) required large amounts of computer analytical time when applied to stock identification problems with a baseline as comprehensive as employed in the current study. Given this limitation, a new version of the program was developed by our laboratory as a C-based program which is available from the Molecular Genetics Laboratory website (http://www-sci.pac.dfo-mpo.gc.ca/mgl/data_e.htm). In the analysis, ten 20,000-iteration Monte Carlo Markov chains of estimated stock compositions were produced, with initial starting values for each chain set at 0.90 for a particular population which was different for each chain. Estimated stock compositions were considered to have converged when the shrink factor was < 1.2 for the 10 chains (Pella and Masuda 2001). The last 1,000 iterations from each of the 10 chains were then combined, and for each fish the probability of originating from each population in the baseline was determined. These individual probabilities were summed over all fish in the sample, and divided by the number of fish sampled to provide the point estimate of stock composition. Standard deviations of estimated stock compositions were determined from the last 1,000 iterations from each of the 10 chains incorporated in the analysis.

Results and Discussion

Chum Salmon

On a seasonal basis, chum salmon spawning populations from the White River drainage accounted for 55% of the run, whereas, mainstem spawning Yukon River populations were estimated to comprise 41% of the chum salmon migrating past the Bio Island fish wheels (Table 2). Approximately 1% of the chum salmon sampled were estimated to be from early-returning populations such as the Chandindu River. Teslin River chum salmon contributed 3% to the total run. Mainstem spawning and White River populations were present throughout the sampling period and, as would be expected, early-returning chum salmon were most prevalent in the earlier sampling periods. Teslin River chum salmon were most prevalent in the last half of the run.

Chinook Salmon

On a seasonal basis, stocks which tended to migrate earlier in the run included: Chinook salmon from the Teslin River drainage which accounted for about 13% of total 2006 run; and Chinook salmon from the north Yukon mainstem tributaries (Klondike River, Chandindu River, Fifty mile) which were estimated to have comprised 10% of the returning Chinook salmon. Stocks which exhibited early to middle timing included: Stewart River Chinook salmon which were estimated to have comprised 13% of the total run; and, Chinook salmon of Pelly River origin which were estimated to have comprised about 12% of the total run. Chinook salmon from the Carmacks area tributaries (Big Salmon, North Big Salmon, Little Salmon, Tatchun Creek) were most common in the middle and later portion of the run, and accounted for about 33% of total run. Mid-Yukon mainstem and Nordenskiold River fish were most common in the later portion of the run, and comprised about 10% of the total number of fish sampled. Upper Yukon River tributaries accounted for about 6% of the total run, with migration timing of these populations also in the later portion of the run. Finally, White River Chinook salmon were estimated to have comprised only about 1% of the run (Table 3).

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Table 1. Baseline used to estimate stock compositions of chum and Chinook salmon from the fish wheel tagging program at Bio Island, 2006.

Region	Populations
Chum salmon	
Mainstem Yukon River	Mainstem Yukon at Pelly River, Tatchun Creek, Big Creek and Minto
White River	Kluane River, Donjek River
Teslin River	Teslin River
Yukon early	Chandindu River
Chinook salmon	
Upper Yukon tributaries	Wolf Creek, Michie Creek, Whitehorse hatchery, Takhini River
Teslin River	Morley River, Gladys Creek, Nisutlin River
Carmacks Area tributaries	Little Salmon, Big Salmon, North Big Salmon, Tatchun Creek
Mid-mainstem/Nordenskiold	Mainstem Yukon River, Nordenskiold River, Minto
Pelly River	Big Kalzas River, Little Kalzas River, Blind Creek, Earn River, Glenlyon, Pelly River
Stewart River	Mayo River, Stewart River
North Yukon mainstem tributaries	Chandindu River, Klondike River, Fifty mile
White River	Tincup Creek

Table 2. Estimated percentage stock composition of chum salmon migrating past the fish wheel tagging program at Bio Island, 2006.

Stock compositions were estimated using 14 microsatellite loci and the baseline outlined in Table 1. Standard deviations of the estimates are in parentheses.

Region	Weekly Sampling															
	Aug 6-26 Sample size 16		Aug 27-Sept 2 25		Sept 3-9 63		Sept 10-16 120		Sept 17-23 267		Sept 24-30 175		Oct1-7 62		Seasonal 728	
Tatchun	3.5	(7.0)	0.2	(1.7)	1.8	(3.8)	3.9	(4.0)	3.7	(3.3)	6.4	(6.2)	4.9	(6.5)	0.4	(0.9)
Pelly	0.4	(2.8)	2.2	(5.6)	15.4	(6.3)	1.4	(1.9)	0.1	(0.4)	1.3	(1.5)	3.5	(4.7)	0.7	(0.8)
Big Creek	24.5	(12.0)	23.5	(10.7)	17.5	(6.9)	22.8	(5.2)	33.2	(4.7)	26.0	(6.6)	35.5	(9.7)	35.3	(2.4)
Minto	0.7	(3.6)	0.6	(2.4)	0.7	(2.1)	4.1	(3.0)	7.6	(3.0)	10.9	(4.5)	0.5	(1.8)	4.7	(1.5)
Kluane	1.5	(4.6)	51.3	(19.4)	43.0	(9.4)	25.3	(7.7)	13.4	(4.2)	22.0	(5.9)	27.1	(7.2)	0.3	(0.5)
Donjek	41.3	(12.3)	21.6	(18.6)	20.9	(8.9)	41.5	(8.1)	37.4	(4.7)	29.0	(6.4)	21.9	(6.9)	54.6	(2.0)
Teslin	0.1	(2.2)	0.2	(1.8)	0.0	(0.6)	0.9	(1.3)	4.5	(1.6)	4.5	(2.2)	6.5	(4.4)	3.1	(0.9)
Chandindu	28.1	(10.8)	0.2	(1.7)	0.6	(1.6)	0.1	(0.6)	0.1	(0.3)	0.0	(0.2)	0.1	(0.9)	1.0	(0.5)
Canadian mainstem	29.1	(11.7)	26.6	(9.3)	35.4	(6.4)	32.2	(4.7)	44.6	(3.4)	44.6	(4.3)	44.5	(7.5)	41.0	(2.0)
White River	42.7	(11.7)	72.9	(9.3)	63.9	(6.4)	66.8	(4.6)	50.7	(3.2)	51.0	(4.0)	49.0	(6.6)	54.9	(1.9)
Teslin River	0.1	(2.2)	0.2	(1.8)	0.0	(0.6)	0.9	(1.3)	4.5	(1.6)	4.5	(2.2)	6.5	(4.4)	3.1	(0.9)
Yukon Early	28.1	(10.8)	0.2	(1.7)	0.6	(1.6)	0.1	(0.6)	0.1	(0.3)	0.0	(0.2)	0.1	(0.9)	1.0	(0.5)

Table 3. Estimated percentage stock composition of Chinook salmon migrating past the fish wheel tagging program at Bio Island, 2006. Stock compositions were estimated using 13 microsatellite loci and the baseline outlined in Table 1. Standard deviations of the estimates are in parentheses.

Sample size	Weekly Sampling											
	July-06 49		July-17 103		July-23 215		July-30 231		Aug-06 149		Seasonal 747	
Big Salmon	3.8	(4.9)	1.0	(2.5)	10.2	(3.2)	9.5	(3.5)	11.2	(4.2)	9.4	(2.3)
L. Salmon	0.2	(1.3)	0.6	(1.7)	17.5	(3.9)	23.5	(4.6)	11.5	(5.7)	14.9	(2.7)
N Big Salmon	0.7	(2.0)	0.3	(1.2)	0.4	(1.1)	0.1	(0.5)	0.1	(0.5)	0.3	(0.6)
Tatchun	1.4	(2.5)	0.1	(0.5)	4.3	(1.9)	7.4	(2.9)	29.2	(5.3)	8.4	(1.5)
Chandindu Riv	43.0	(7.5)	22.2	(5.1)	6.1	(2.2)	1.4	(0.9)	0.0	(0.2)	7.0	(1.2)
Fifty-Mile	0.0	(0.5)	0.1	(0.7)	2.2	(2.0)	5.4	(3.2)	1.2	(2.9)	1.4	(1.2)
Klondike	0.5	(2.3)	3.0	(3.7)	1.6	(1.8)	0.1	(0.4)	0.1	(0.3)	1.9	(1.0)
Minto	0.1	(0.6)	0.1	(0.6)	0.1	(0.4)	0.5	(1.4)	0.3	(1.0)	0.3	(0.4)
Nordenskiold	0.0	(0.4)	0.8	(1.1)	0.0	(0.1)	0.1	(0.3)	0.0	(0.3)	0.1	(0.2)
Yukon main	0.3	(1.6)	0.3	(1.1)	4.0	(3.0)	16.1	(4.0)	20.3	(6.8)	9.9	(1.9)
Big Kalzas	8.4	(5.3)	0.6	(1.3)	0.0	(0.2)	0.0	(0.1)	0.1	(0.5)	0.0	(0.2)
Blind Cr	0.6	(2.2)	13.2	(4.8)	3.5	(2.3)	0.4	(1.0)	0.4	(1.2)	3.5	(1.2)
Earn River	0.3	(1.4)	1.3	(2.5)	1.4	(1.6)	0.0	(0.2)	2.1	(2.3)	0.7	(0.9)
Glenlyon	3.6	(3.5)	0.1	(0.6)	0.3	(0.8)	0.0	(0.2)	0.7	(1.0)	0.3	(0.5)
Little Kalzas	0.2	(1.3)	0.4	(1.5)	0.9	(1.3)	2.9	(1.3)	1.2	(1.4)	2.2	(0.8)
Pelly	4.9	(7.5)	8.5	(7.0)	9.5	(3.2)	0.1	(0.4)	2.6	(3.7)	5.6	(1.8)
Mayo	14.5	(7.2)	14.0	(7.2)	8.9	(3.8)	6.5	(3.8)	8.9	(3.8)	9.6	(2.1)
Stewart	1.8	(3.8)	5.3	(4.3)	6.7	(2.8)	0.1	(0.4)	0.3	(0.9)	3.8	(1.4)
Gladys	0.6	(1.9)	10.7	(5.7)	4.0	(3.0)	3.3	(2.4)	0.6	(2.2)	3.0	(1.1)
Morley	1.2	(2.9)	6.4	(4.7)	4.8	(2.8)	3.2	(2.8)	1.9	(2.0)	4.7	(1.9)
Nisutlin	13.7	(6.0)	8.6	(5.4)	3.9	(2.1)	6.0	(2.8)	2.9	(2.4)	5.3	(1.9)
Michie	0.0	(0.5)	0.8	(1.1)	0.0	(0.2)	0.2	(0.7)	0.1	(0.4)	0.1	(0.3)
Takhini	0.1	(0.7)	0.0	(0.2)	4.5	(1.7)	3.8	(1.7)	1.3	(1.4)	2.2	(0.7)
Whitehorse	0.0	(0.4)	0.1	(0.5)	1.8	(1.0)	7.5	(2.0)	2.8	(1.8)	3.6	(0.8)
Wolf	0.0	(0.4)	0.0	(0.4)	0.1	(0.4)	0.4	(1.0)	0.1	(0.6)	0.1	(0.3)
Tincup	0.0	(0.4)	1.5	(2.0)	3.3	(1.5)	1.3	(1.0)	0.0	(0.2)	1.7	(0.6)
Carmacks tribs	6.1	(5.1)	1.9	(3.2)	32.4	(4.7)	40.5	(4.7)	52.0	(7.2)	33.0	(2.9)
Lower Yukon tribs	43.5	(7.2)	25.3	(4.7)	9.9	(2.9)	6.9	(3.3)	1.3	(3.0)	10.3	(1.7)
Yukon main/Norden	0.4	(1.8)	1.2	(1.7)	4.1	(3.1)	16.7	(4.3)	20.7	(6.8)	10.2	(2.0)
Pelly River	18.0	(7.3)	24.0	(6.4)	15.6	(3.5)	3.5	(1.7)	7.2	(3.4)	12.4	(1.9)
Stewart River	16.4	(7.5)	19.4	(7.0)	15.6	(4.0)	6.5	(3.8)	9.1	(3.8)	13.4	(2.2)
Teslin River	15.5	(5.8)	25.7	(6.6)	12.7	(4.2)	12.5	(3.5)	5.4	(3.3)	13.0	(1.9)
Upper Yukon tribs	0.1	(1.0)	1.0	(1.2)	6.5	(1.9)	12.0	(2.6)	4.3	(2.2)	6.1	(1.0)
White River	0.0	(0.4)	1.5	(2.0)	3.3	(1.5)	1.3	(1.0)	0.0	(0.2)	1.7	(0.6)