

*Pacific Salmon Commission*  
*Northern Fund*

**Northern Boundary Area Sockeye Salmon Genetic Stock Identification  
For Year 2017 District 101 Gillnet and District 104 Purse Seine Fisheries**

FINAL Report

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## INTRODUCTION

Provisions outlined in Chapter 2 of the Pacific Salmon Treaty specify harvest sharing arrangements of Nass and Skeena River sockeye salmon (*Oncorhynchus nerka*) between the United States and Canada. This treaty allows the United States to harvest a fixed percentage, averaged over ten years, of the annual allowable harvest (AAH) of Nass sockeye in the Alaskan District 101 gillnet fishery (GNF) and of Nass and Skeena sockeye in the District 104 purse seine fishery (PSF) prior to Statistical Week 31 (late July). There is also a District 101 PSF, but the catch in this fishery is not limited by the annex; it is used however in calculating the total return of Alaska, Nass and Skeena River stocks (along with districts 102, 103 seine and 106 gillnet). Figure 1 illustrates the locations of the Alaska Department of Fish and Game (ADF&G) commercial fishing districts in the Northern Boundary area.

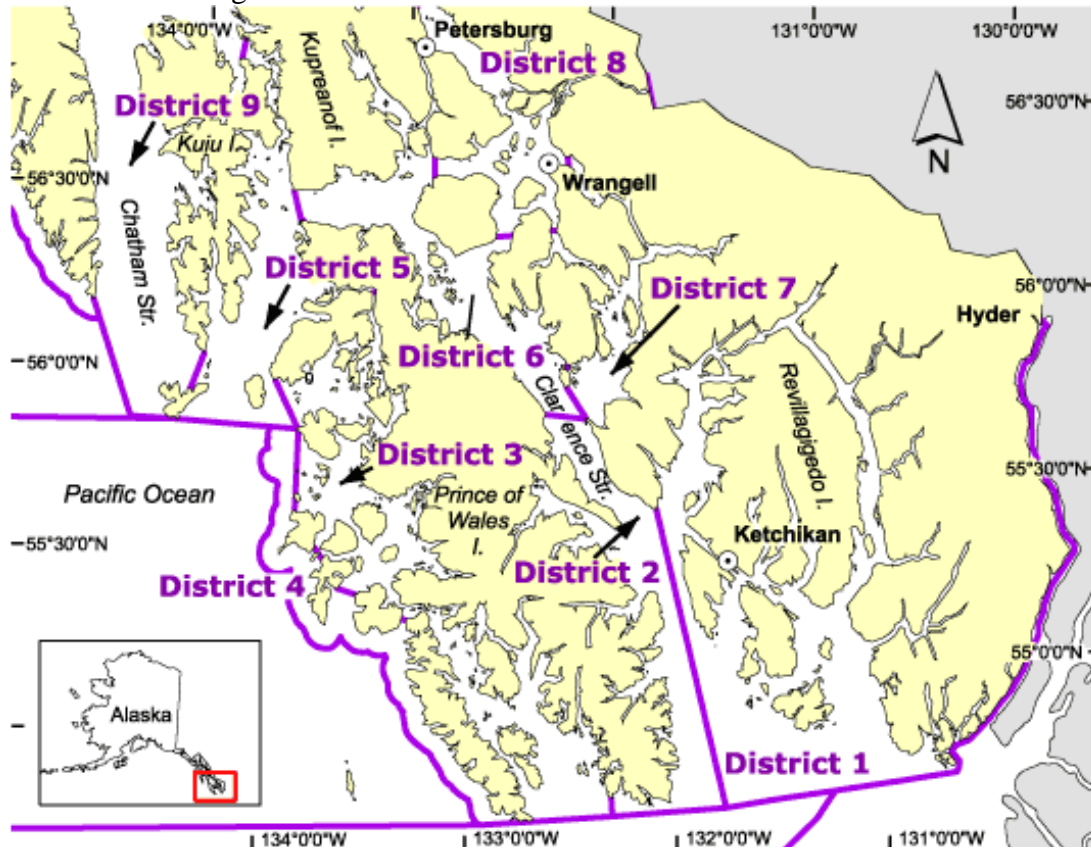
Accurate estimates of the stock composition of sockeye salmon caught in boundary area gillnet and purse seine fisheries (few are caught in troll fisheries) are required to estimate the total return (catch plus escapement) of stocks subject to harvest sharing agreements. The estimated total return is then used in calculating the percentage of the AAH caught in the District 101 gillnet and District 104 purse seine fisheries. The AAH is calculated over the ten-year annex period. This approach allows for traditional fishing patterns based on stock abundance, recognizing that for some years more fish would be caught which would be compensated by other years in which less would be harvested.

It has been recognized for some time that U.S. and Canadian fishermen intercept salmon originating from the other country. Initial studies investigating the stock origins of pink (*O. gorbuscha*) and sockeye salmon caught in the Northern Boundary region between Alaska and British Columbia used mark-recapture techniques (Pella et al., 1993). These techniques involved tagging fish caught in boundary fisheries and recapturing them at various weirs and other in-river escapement enumeration projects. This study found that a significant percent of the fish caught in districts 101 and 104 originated from Canadian stocks (Pella et al., 1993). While informative, these tagging experiments were relatively expensive and labor intensive.

A study was undertaken in 1982 to evaluate scale pattern analysis (SPA) as a means to discriminate particular stocks of fish (Marshall, 1984). This important study showed that sockeye salmon in the Alaska-British Columbia Northern Boundary area could be accurately discriminated using scales. SPA was used by ADF&G to determine stock proportions for sockeye salmon caught in the commercial sockeye fisheries in districts 101 and 104 until 2012.

While effective, SPA required yearly examination of source populations for each of the four major age classes (1.2, 1.3, 2.2 and 2.3) since the scale baseline patterns are strongly affected by varying environmental conditions. The requirement to reestablish or revalidate the scale pattern baseline was expensive and burdensome. The use of more stable markers has eliminated this necessity. Like scale patterns, DNA patterns can also be used to discriminate stocks of salmon (Milner et al., 1985). Given that salmon return to their natal streams with high fidelity, they represent naturally occurring isolated populations in which genetic allele frequencies can change due to the isolation and adaptation of particular populations. These changes in allele frequencies can then be used to distinguish salmon stocks to a finer degree of resolution than SPA. For example,

scale analysis can efficiently separate 4 large stock groups (Alaska, Nass, Skeena and Fraser) whereas genetic analysis can separate 7 stock groups, adding to the knowledge available to manage area fisheries.



**Figure 1.** Geographic location of ADF&G commercial fishing districts 101 (labeled District 1) and 104 (labeled District 4). Map obtained from the ADF&G web page (<http://www.cfdg.state.ak.us/region1/finfish/salmon/maps/ketchikan.php>).

Allozymes are naturally occurring protein variants which have been used as genetic markers. As part of a study to estimate stock composition of sockeye salmon harvested in the 1987 Northern Boundary sockeye fisheries in districts 104 and 106 (Pella et al., 1998), four markers were used which included two unlinked allozyme markers (*PGM-1\** and *PGM-2\**), freshwater age, and a brain-tissue parasite (*Myxobolus arcticus*). Freshwater age and pathogen exposure are traits that can be used in combination with other markers to infer the stock composition of mixtures (Fournier et al., 1984; Pella and Milner, 1987). The 1987 study provided estimated proportions of 13 stock groups in the District 104 fisheries and confirmed that the majority of sockeye salmon caught were of Canadian origin (Pella et al., 1998). This analysis demonstrated that genetic markers could be effective in estimating the stock composition of sockeye salmon caught in Northern Boundary fisheries.

Although allozymes have been used in many genetic studies in salmon, it can be laborious to complete all the lab methods necessary to score them. Since then, additional genetic markers have been evaluated including microsatellite DNA repeats and single

**Table 1. Sockeye salmon baseline populations used in analysis.**

| <i>Pop. #</i> | <i>Description</i>           | <i>Region</i> | <i>Pop. #</i> | <i>Description</i>              | <i>Region</i> |
|---------------|------------------------------|---------------|---------------|---------------------------------|---------------|
| 1             | Bainbridge Lake              | 1             | 44            | Dangerous River                 | 1             |
| 2             | Coghill Lake                 | 1             | 45            | Akwe River                      | 1             |
| 3             | Eshamy Creek                 | 1             | 46            | East Alsek River                | 1             |
| 4             | Main Bay                     | 1             | 47            | Datlasaka Creek                 | 5             |
| 5             | Miners Lake                  | 1             | 48            | Goat Creek                      | 5             |
| 6             | Eyak Lake - Middle Arm       | 1             | 49            | Border Slough 2007 & 2008       | 5             |
| 7             | Eyak Lake - South beaches    | 1             | 50            | Border Slough 2009 & 2011       | 5             |
| 8             | Eyak Lake - Hatchery Creek   | 1             | 51            | Tweedsmuir 2007                 | 5             |
| 9             | Mendeltna Creek              | 1             | 52            | Tweedsmuir 2009                 | 5             |
| 10            | Swede Lake                   | 1             | 53            | Vern Ritchie                    | 5             |
| 11            | East Fork Gulkana River      | 1             | 54            | Neskataheen Lake                | 5             |
| 12            | Gulkana River - East Fork    | 1             | 55            | Klukshu River 2006              | 5             |
| 13            | Paxon Lake                   | 1             | 56            | Klukshu River 2007              | 5             |
| 14            | Mentasta Lake                | 1             | 57            | Kudwat Creek                    | 5             |
| 15            | Tanada Creek                 | 1             | 58            | Tatshenshini - Bridge/Silver    | 5             |
| 16            | Tanada Lake - lower outlet   | 1             | 59            | Tatshenshini - Stinky Creek     | 5             |
| 17            | Tanada Lake - shore          | 1             | 60            | Upper Tatshenshini              | 5             |
| 18            | Klutina River                | 1             | 61            | Little Tatshenshini Lake        | 5             |
| 19            | Klutina Lake                 | 1             | 62            | Kwatini River                   | 5             |
| 20            | Bear Hole - Klutina          | 1             | 63            | Blanchard River 2007            | 5             |
| 21            | Banana Lake - Klutina        | 1             | 64            | Blanchard River 2009            | 5             |
| 22            | St. Anne Creek               | 1             | 65            | Bear Flats - Chilkat            | 1             |
| 23            | Mahlo River                  | 1             | 66            | Mule Meadows - Chilkat          | 1             |
| 24            | Tonsina Lake                 | 1             | 67            | Mosquito Lake - Chilkat         | 1             |
| 25            | Long Lake                    | 1             | 68            | Chilkat Lake 2007               | 1             |
| 26            | Tebay River                  | 1             | 69            | Chilkat Lake 2013               | 1             |
| 27            | Steamboat Lake - Bremner     | 1             | 70            | Chilkoot River                  | 1             |
| 28            | Salmon Creek - Bremner       | 1             | 71            | Chilkoot Lake - Bear Creek      | 1             |
| 29            | Clear Creek                  | 1             | 72            | Chilkoot Lake - beaches         | 1             |
| 30            | McKinley Lake 2007           | 1             | 73            | Vivid Lake                      | 1             |
| 31            | McKinley Lake 2008           | 1             | 74            | Seclusion Lake                  | 1             |
| 32            | McKinley Lake 1991           | 1             | 75            | North Berg Bay Inlet Creek 1991 | 1             |
| 33            | McKinley Lake - Salmon Creek | 1             | 76            | North Berg Bay Inlet Creek 1992 | 1             |
| 34            | Martin Lake                  | 1             | 77            | Bartlett River                  | 1             |
| 35            | Martin River Slough          | 1             | 78            | Neva Lake 2008                  | 1             |
| 36            | Tokun Lake                   | 1             | 79            | Neva Lake 2009 & 2013           | 1             |
| 37            | Bering Lake                  | 1             | 80            | Hoktaheen - main inlet          | 1             |
| 38            | Kushtaka Lake                | 1             | 81            | Hoktaheen - outlet              | 1             |
| 39            | Mountain Stream              | 1             | 82            | Hoktaheen - marine waters       | 1             |
| 40            | Situk Lake                   | 1             | 83            | Klag Bay Stream                 | 1             |
| 41            | Old Situk River              | 1             | 84            | Ford Arm Lake                   | 1             |
| 42            | Lost/Tahwah Rivers           | 1             | 85            | Ford Arm Creek                  | 1             |
| 43            | Ahmklin River                | 1             | 86            | Redoubt Lake                    | 1             |

**Table 1.** continued.

| <i>Pop. #</i> | <i>Description</i>              | <i>Region</i> | <i>Pop. #</i> | <i>Description</i>            | <i>Region</i> |
|---------------|---------------------------------|---------------|---------------|-------------------------------|---------------|
| 87            | Salmon Lake                     | 1             | 130           | Andy Smith slough             | 5             |
| 88            | Benzeman Lake                   | 1             | 131           | Porcupine                     | 5             |
| 89            | Falls Lake                      | 1             | 132           | Devil's Elbow 2007 & 2008     | 5             |
| 90            | Redfish Lake                    | 1             | 133           | Devil's Elbow 2009            | 5             |
| 91            | Kutlaku 2003                    | 1             | 134           | Scud River                    | 5             |
| 92            | Kutlaku 2012                    | 1             | 135           | Iskut River                   | 5             |
| 93            | Kutlaku 2013                    | 1             | 136           | Iskut River (Craigson Slough) | 5             |
| 94            | Lace River                      | 1             | 137           | Craig River-CAN               | 5             |
| 95            | Berners Bay                     | 1             | 138           | Bronson Slough                | 5             |
| 96            | Antler-Gilkey River             | 1             | 139           | Shakes Slough                 | 5             |
| 97            | Windfall Lake                   | 1             | 140           | Christina Lake                | 5             |
| 98            | Steep Creek                     | 1             | 141           | Petersburg Lake               | 1             |
| 99            | Lake Creek (Auke Creek Weir)    | 1             | 142           | Kah Sheets Lake               | 1             |
| 100           | Crescent Lake                   | 1             | 143           | Mill Creek Weir Early         | 1             |
| 101           | Speel Lake                      | 1             | 144           | Mill Creek Weir Late          | 1             |
| 102           | Snettisham Hatchery 2006 & 2007 | 1             | 145           | Kunk Lake                     | 1             |
| 103           | Snettisham Hatchery 2013        | 1             | 146           | Thoms Lake                    | 1             |
| 104           | Pavlof River                    | 1             | 147           | Red Bay Lake                  | 1             |
| 105           | Kook Lake Late                  | 1             | 148           | Salmon Bay Lake               | 1             |
| 106           | Kook Lake early                 | 1             | 149           | Shiple Lake                   | 1             |
| 107           | Sitkoh Lake                     | 1             | 150           | Sarkar Lakes                  | 1             |
| 108           | Lake Eva                        | 1             | 151           | Hatchery Creek                | 1             |
| 109           | Hasselborg Lake                 | 1             | 152           | Luck Lake                     | 1             |
| 110           | Kanalku Lake                    | 1             | 153           | Big Lake                      | 1             |
| 111           | Kuthai Lake                     | 5             | 154           | McDonald Lake                 | 4             |
| 112           | King Salmon Lake                | 5             | 155           | Karta River                   | 1             |
| 113           | Little Trapper Lake             | 5             | 156           | Unuk River 2007               | 1             |
| 114           | Little Tatsamenie 2011          | 5             | 157           | Unuk River 2008               | 1             |
| 115           | Tatsamenie Lake                 | 5             | 158           | Helm Lake                     | 1             |
| 116           | Tahltan Lake90                  | 5             | 159           | Heckman Lake                  | 1             |
| 117           | Tahltan Lake06                  | 5             | 160           | Mahoney Creek                 | 1             |
| 118           | Hackett River                   | 5             | 161           | Kegan Lake                    | 1             |
| 119           | Nahlin River                    | 5             | 162           | Fillmore Lake                 | 1             |
| 120           | Taku River                      | 5             | 163           | Klawock - Three Mile          | 1             |
| 121           | Taku Mainstem - Takwahoni/Sinwa | 5             | 164           | Klawock - Inlet Creek         | 1             |
| 122           | Shustahini Slough               | 5             | 165           | Hetta Lake                    | 1             |
| 123           | Tuskwa/Chunk Slough             | 5             | 166           | Hetta Creek - middle run      | 1             |
| 124           | Yellow Bluff Slough             | 5             | 167           | Hetta Creek - early run       | 1             |
| 125           | Tulsequah River                 | 5             | 168           | Eek Creek                     | 1             |
| 126           | Fish Creek                      | 5             | 169           | Klakas Lake                   | 1             |
| 127           | Yehring Creek                   | 5             | 170           | Essowah Lake                  | 1             |
| 128           | Chutine River                   | 5             | 171           | Hugh Smith Lake               | 1             |
| 129           | Chutine Lake                    | 5             | 172           | Hugh Smith - Buschmann Creek  | 1             |



**Table 1. continued.**

| <i>Pop. #</i> | <i>Description</i>            | <i>Region</i> | <i>Pop. #</i> | <i>Description</i>              | <i>Region</i> |
|---------------|-------------------------------|---------------|---------------|---------------------------------|---------------|
| 173           | Hugh Smith - Cobb Creek       | 1             | 216           | Raft River                      | 6             |
| 174           | Kwinageese                    | 2             | 217           | Adams River                     | 6             |
| 175           | Bowser Lake                   | 2             | 218           | Middle Shuswap River            | 6             |
| 176           | Bonney Creek                  | 2             | 219           | Scotch River                    | 6             |
| 177           | Damdochax Creek               | 2             | 220           | Gates Creek                     | 6             |
| 178           | Meziadin Lake                 | 2             | 221           | Birkenhead River                | 6             |
| 179           | Hanna Creek                   | 2             | 222           | Weaver Creek                    | 6             |
| 180           | Tintina Creek                 | 2             | 223           | Harrison River                  | 6             |
| 181           | Gingit Creek                  | 2             | 224           | North Thompson                  | 6             |
| 182           | Alastair Lake                 | 3             | 225           | Naden River                     | 7             |
| 183           | Lakeelse Lake                 | 3             | 226           | QCI - Yakoun Lake               | 7             |
| 184           | Sustut River                  | 3             | 227           | Kitimat River                   | 7             |
| 185           | Salix Bear                    | 3             | 228           | Bloomfield Lake                 | 7             |
| 186           | Motase Lake                   | 3             | 229           | Tankeeah River 2003             | 7             |
| 187           | Slamgeesh River               | 3             | 230           | Tankeeah River 2005             | 7             |
| 188           | Babine River                  | 3             | 231           | Central Coast - Amback Creek    | 7             |
| 189           | Four Mile Creek               | 3             | 232           | Kitlope Lake                    | 7             |
| 190           | Pinkut Creek                  | 3             | 233           | Great Central Lake              | 7             |
| 191           | Grizzly Creek                 | 3             | 234           | Vancouver Island - Quatse River | 7             |
| 192           | Pierre Creek                  | 3             | 235           | Okanagan River                  | 7             |
| 193           | Fulton River                  | 3             | 236           | Lake Pleasant                   | 7             |
| 194           | Morrison                      | 3             | 237           | Issaquah Creek                  | 7             |
| 195           | Lower Tahlo River             | 3             | 238           | Lake Wenatchee                  | 7             |
| 196           | Tahlo Creek                   | 3             |               |                                 |               |
| 197           | McDonell Lake (Zymoetz River) | 3             |               |                                 |               |
| 198           | Kitsumkalum Lake 2006         | 3             |               |                                 |               |
| 199           | Kitsumkalum Lake 2012         | 3             |               |                                 |               |
| 200           | Kitwanga River                | 3             |               |                                 |               |
| 201           | Stephens Creek                | 3             |               |                                 |               |
| 202           | Nangeese River                | 3             |               |                                 |               |
| 203           | Kispiox River                 | 2             |               |                                 |               |
| 204           | Swan Lake                     | 3             |               |                                 |               |
| 205           | Nanika River                  | 3             |               |                                 |               |
| 206           | Trembleur - Kynock            | 6             |               |                                 |               |
| 207           | Tachie River                  | 6             |               |                                 |               |
| 208           | Stellako River                | 6             |               |                                 |               |
| 209           | Fraser Lake                   | 6             |               |                                 |               |
| 210           | Mitchell River                | 6             |               |                                 |               |
| 211           | Horsefly River                | 6             |               |                                 |               |
| 212           | Nahatlatch River              | 6             |               |                                 |               |
| 213           | Cultus Lake                   | 6             |               |                                 |               |
| 214           | Chilliwack Lake               | 6             |               |                                 |               |
| 215           | Chilko Lake                   | 6             |               |                                 |               |

nucleotide polymorphisms (SNPs). Like allozymes, both microsatellite and SNP markers can efficiently be used to separate stocks of salmon (Beacham et al., 2008; Habicht et al., 2004, 2010; Smith et al., 2005a). While Canadian scientists use microsatellite markers for many of their Northern Boundary studies, ADF&G uses SNPs. Numerous studies have been completed outlining the advantages and disadvantages of each, although both have the resolving power necessary to accurately perform stock composition studies (Smith et al., 2007).

ADF&G has developed a sockeye new SNP baseline with 48 SNP markers (Habicht et al. 2007, 2010, Dann et al., 2012). This baseline replaces the 43 was used by ADF&G in 2004 and 2005; and by NOAA/NMFS/Alaska Fishery Science Center/Auke Bay Laboratories (ABL) in 2006-15 (Guthrie et al. 2009-17) for genetic stock composition analyses for districts 101 and 104. Previously, 84 sockeye populations were part of the SNP baseline, but in the new baseline that number has increased to 171 in 2016 and 238 (Table 1) in 2017 to fulfill a request of the Northern Boundary Technical Committee (NBTC) for additional baseline populations from Canada. As part of this process, the resolving power of the SNP baseline was evaluated using simulated mixture analyses, and this baseline was shown to be fully capable of distinguishing 7 Northern Boundary sockeye stock groups which are more relevant to the run reconstruction used by the Pacific Salmon Commission (PSC) than the 13 stock groups used previously (Table 2) (Oliver 2009).

**Table 2.** Regional grouping of populations for stock composition analysis.

| <i>Region</i> | <i>Area</i>         |
|---------------|---------------------|
| 1             | Alaska              |
| 2             | Nass River          |
| 3             | Skeena River        |
| 4             | McDonald Lake       |
| 5             | Transboundry Rivers |
| 6             | Fraser River        |
| 7             | South Migrating     |

Problems in accurately estimating stock proportions of catches and total returns of sockeye salmon in the early years of the Pacific Salmon Treaty resulted in an extensive investigation of run reconstruction modeling by the bilateral NBTC. The NBTC concluded that improved stock identification techniques are needed for run reconstruction models. As opposed to SPA, genetic techniques have the advantage of a relatively stable baseline (does not change yearly) and the analysis can be highly automated. Congruence was found between the two techniques, so genetic analysis replaced SPA for estimating stock composition of sockeye salmon caught in Northern Boundary fisheries in 2012. A blind testing study performed determined genetic markers are the viable method to replace SPA (Oliver personal communication, 2011).

## OBJECTIVE

The purpose of this study was to genetically analyze axillary process (AXP) samples from 4,292 sockeye salmon harvested in the 2017 District 101 gillnet and District 104 purse seine sockeye fisheries to determine proportions of Canadian and U.S. fish. A SNP genetic baseline of 48 SNPs assayed in 238 sockeye populations from southeast Alaska and British Columbia, and Washington was developed by ADF&G (Habicht et al., 2010, Dann et al., 2012).

**Table 3.** 48 SNP assays used to discriminate Northern Boundary sockeye populations.

| #  | Name                           | #  | Name                                |
|----|--------------------------------|----|-------------------------------------|
| 1  | <i>One_agt-132</i>             | 25 | <i>One_STR07</i>                    |
| 2  | <i>One_apoe-83</i>             | 26 | <i>One_SUMO1-6</i>                  |
| 3  | <i>One_CFP1</i>                | 27 | <i>One_sys1-230</i>                 |
| 4  | <i>One_cin-177</i>             | 28 | <i>One_taf12-248</i>                |
| 5  | <i>One_CO1 (mitochondrial)</i> | 29 | <i>One_Tf_ex3-182</i>               |
| 6  | <i>One_E2</i>                  | 30 | <i>One_U1003-75</i>                 |
| 7  | <i>One_GHII-2461</i>           | 31 | <i>One_U1004-183</i>                |
| 8  | <i>One_ghsR-66</i>             | 32 | <i>One_U1009-91</i>                 |
| 9  | <i>One_HGFA</i>                | 33 | <i>One_U1010-81</i>                 |
| 10 | <i>One_HpaI-436</i>            | 34 | <i>One_U1013-108</i>                |
| 11 | <i>One_HpaI-99</i>             | 35 | <i>One_U1016-115 (not resolved)</i> |
| 12 | <i>One_IL8r-362</i>            | 36 | <i>One_U1101</i>                    |
| 13 | <i>One_metA-253</i>            | 37 | <i>One_U1201-492</i>                |
| 14 | <i>One_MHC2_190</i>            | 38 | <i>One_U1203-175</i>                |
| 15 | <i>One_Mkpro-129</i>           | 39 | <i>One_U1205-57</i>                 |
| 16 | <i>One_Ots213-181</i>          | 40 | <i>One_U1208-67</i>                 |
| 17 | <i>One_rab1a-76</i>            | 41 | <i>One_U1212-106</i>                |
| 18 | <i>One_RAG3-93</i>             | 42 | <i>One_U1214-107</i>                |
| 19 | <i>One_redd1-414</i>           | 43 | <i>One_U1216-230</i>                |
| 20 | <i>One_RF-112</i>              | 44 | <i>One_U301-92</i>                  |
| 21 | <i>One_spf30-207</i>           | 45 | <i>One_U503-170</i>                 |
| 22 | <i>One_srp09-127</i>           | 46 | <i>One_U504-141</i>                 |
| 23 | <i>One_ssrd-135</i>            | 47 | <i>One_vatf-214</i>                 |
| 24 | <i>One_STC-410</i>             | 48 | <i>One_zP3b</i>                     |

## METHODS

### *Genetic baseline and population grouping*

Genetic samples from 238 baseline populations (Table 1) were collected by ADF&G in collaboration with many other laboratories including ABL and the Canadian Department of Fisheries and Oceans. The 238 populations were grouped into 7 regions (Table 2) based on manager needs, the PSC groupings, geographical location, and historical knowledge.

### *Sample Collection*

Matched genetic and scale samples were collected by port samplers and observers from ADF&G. Samples were collected from the District 101 GNF and from the District

104 PSF. Genetic samples were clipped AXP that were dried and stored on Whatman™ paper. The genetic samples were shipped to ABL for analysis and stored at room temperature. ADF&G collected genetic and scale samples from a maximum of 310 (Table 4&5) fish per statistical week for each district, of which over 99% were successfully analyzed (Table 4&5).

#### *DNA Extraction*

DNA was extracted from the AXP into 96-well plates with the QIAGEN DNeasy Blood and Tissue Kits as described by the manufacturer (QIAGEN, Inc.). In brief, small pieces of tissue (~20 mg) were excised from Whatman™ stored axillary processes. The tissue pieces were digested in a proteinase solution for at least 3 hours at 55°C. Protease digestions were performed in 96 well plates. After digestion, the samples were purified with either QIAxtractor or Corbett X-tractor robot producing eluted DNA which was stored at -20 °C.

#### *Single Nucleotide Polymorphism (SNP) Analysis*

SNP genotyping was performed using Taqman™ chemistries from Life Technologies for 48 previously identified sockeye SNP probes. Of the 48 sockeye SNP markers (Table 3) (Elfstrom et al., 2006; Smith et al., 2005b; Habicht et al., 2007, 2010; Dann et al., 2012), 47 were assayed in this analysis. The remaining assay, *One\_U1016-115* was excluded due to poor resolution. Taqman™ reactions were performed by transferring 1  $\mu$ l of a 1:10 dilution of the eluted purified DNA to wells of a 384 well plate. Four wells were reserved for non-template controls. Each Taqman™ reaction was conducted in a 5  $\mu$ l volume containing the template DNA, Taqman™ Universal PCR Mastermix, No AmpErase UNG (Life Technologies), 900 nm of each PCR primer, and 200 nm probe. Thermal cycling was performed on an ABI Dual 384-Well GeneAmp PCR System 9700 using the protocol from Habicht et al. (2010).

#### *Allele Scoring*

After amplification, the Taqman™ genotyping reactions were assayed on a Life Technologies QuantStudio and scored using QuantStudio 12K Flex Software v1.2.2. Individual genotypes were imported into our genetic database developed with Progeny software (Progeny, Inc.).

#### *Mixture Analysis*

A mixture analysis using a Bayesian estimation method (Pella and Masuda, 2001) was implemented using BAYES software and was performed for each weekly mixture sample and each district. For each BAYES analysis, 7 Monte Carlo chains starting at disparate values of stock proportions were configured such that 95% of the stocks came from one designated region with weights equally distributed among the stocks of that region. The remaining 5% was equally distributed among remaining stocks from all other regions. For all estimates, a flat prior of 0.004202 (calculated as 1/238) was used for all 238 populations. Convergence of chains to posterior distributions of stock proportions was determined with Gelman and Rubin shrink factors (Gelman and Rubin 1992), and the first one-half of chains was discarded as burn-in before summarizing

posterior distributions. Most Monte Carlo chain lengths were 10,000; two chain lengths were 100,000 to obtain convergence.

**Table 4.** Sockeye salmon harvested, genetic sample size, genotyping success rate, and percent catch analyzed in each statistical week in the 2017 District 101 Gillnet fishery.

| <i>District 101 Gillnet</i> |             |                       |                  |                 |                   |                |
|-----------------------------|-------------|-----------------------|------------------|-----------------|-------------------|----------------|
| <i>Week</i>                 | <i>2017</i> | <i>2007-2016 Avg.</i> | <i>Extracted</i> | <i>Analyzed</i> | <i>% Analyzed</i> | <i>% Catch</i> |
| 25                          | 4,898       | 5,510                 | 261              | 259             | 99.2              | 5.3            |
| 26                          | 1,641       | 7,902                 | 260              | 254             | 97.7              | 15.5           |
| 27                          | 2,891       | 9,216                 | 220              | 220             | 100.0             | 7.6            |
| 28                          | 2,373       | 6,684                 | 256              | 252             | 98.4              | 10.6           |
| 29                          | 2,084       | 4,797                 | 260              | 259             | 99.6              | 12.4           |
| 30                          | 1,857       | 5,034                 | 260              | 257             | 98.8              | 13.8           |
| 31                          | 3,273       | 5,647                 | 260              | 259             | 99.6              | 7.9            |
| 32                          | 2,486       | 5,399                 | 258              | 257             | 99.6              | 10.3           |
| 33                          | 1,011       | 3,219                 | 260              | 260             | 100.0             | 25.7           |
| 34                          | 1,035       | 1,430                 | 121              | 121             | 100.0             | 11.7           |
| 35                          | 1,079       | 819                   | 220              | 220             | 100.0             | 20.4           |
| 36                          | 292         | 416                   | 0                | 0               | 0.0               | 0.0            |
| 37                          | 85          | 172                   | 0                | 0               | 0.0               | 0.0            |
| 38                          | 65          | 40                    | 0                | 0               | 0.0               | 0.0            |
| 39                          | 3           | 6                     | 0                | 0               | 0.0               | 0.0            |
| <b>Total Catch</b>          | 25,073      | 56,290                |                  |                 |                   | 10.4           |
| <b>Sampled Catch</b>        | 24,628      | 55,657                | 2,636            | 2,618           | 99.3              | 10.6           |

**Table 5.** Sockeye salmon harvested, genetic sample size, genotyping success rate, and percent catch analyzed in each statistical week in the 2017 District 104 Purse Seine fishery.

| <i>District 104 Purse Seine</i> |             |                       |                  |                 |                   |                |
|---------------------------------|-------------|-----------------------|------------------|-----------------|-------------------|----------------|
| <i>Week</i>                     | <i>2017</i> | <i>2007-2016 Avg.</i> | <i>Extracted</i> | <i>Analyzed</i> | <i>% Analyzed</i> | <i>% Catch</i> |
| 29                              | 7,492       | 16,041                | 260              | 259             | 99.6              | 3.5            |
| 30                              | 4,544       | 21,297                | 0                | 0               | 0.0               | 0.0            |
| 31                              | 19,349      | 66,908                | 310              | 306             | 98.7              | 1.6            |
| 32                              | 16,269      | 90,821                | 260              | 260             | 100.0             | 1.6            |
| 33                              | 9,662       | 48,147                | 300              | 300             | 100.0             | 3.1            |
| 34                              | 20,025      | 24,917                | 300              | 299             | 99.7              | 1.5            |
| 35                              | 19,182      | 12,344                | 250              | 250             | 100.0             | 1.3            |
| 36                              | 1,501       | 331                   | 0                | 0               | 0.0               | 0.0            |
| <b>Total Catch</b>              | 98,024      | 280,805               |                  |                 |                   | 1.7            |
| <b>Analyzed Catch</b>           | 91,979      | 259,178               | 1,680            | 1,674           | 99.6              | 1.8            |

## RESULTS

In 2017, 24,628 sockeye salmon were harvested in District 101 GNF which is less than the 2007 to 2016 average of 55,657 (Table 4). In the District 104 PSF 91,979 fish were harvested in 2017 which is much smaller than the 2007-2016 average of 259,178 (Table 5). Sockeye salmon DNA was isolated (Table 4&5) and genotyped for 47 SNP markers from 4,292 fish in 2017. The data was imported into a Progeny database for

analysis. Samples resolved for at least 42 of the 47 SNPs were included in the analyses (i.e. % analyzed in Tables 4&5).

### *Stock Mixture Proportions*

Weekly mixture samples were analyzed with BAYES software. In all of the analyses, the Gelman and Rubin shrink factors were less than 1.2, indicating convergence of the chains to posterior distributions. Results from this analysis are presented in both graphical form (Figure 2) and Table form (Tables 6&7). Figure 2 graphically illustrates the estimated proportions of sockeye salmon endemic to each of the 7 regions that were harvested in each district and statistical week. Tables 6 and 7 provide the same data shown in Figure 2 in numerical format showing the estimated stock group proportions, standard errors, and 95% credible intervals for the 2017 101 GNF and 104 PSF respectively.

Analysis of the stock proportions of sockeye caught in districts 101 GNF and 104 PSF over varying weeks shows interesting trends (Tables 6&7). For example, the sockeye commercial fishery in the 2017 District 101 GNF harvested a greater proportion of Nass Region fish; with a high of 82% in week 25, and a low of 11% in week 32. Skeena fish were not as abundant as in previous years (Table 8), with a high of 45% in week 36, and a low of 2% in week 28. Alaska fish were sporadically abundant with a high of 69% in week 28 and a low of 9% in week 25.

The sockeye commercial fishery in the 2017 District 104 PSF harvested a greater proportion of Skeena River fish ranging from 33% to 54% in weeks 29 through 35. Nass fish were present early at 15% in week 28, while some Fraser fish appearing after week 29 with a high of 39% in week 34. Alaska fish were present throughout with a high of 31% in week 28, and a low of 9% in week 33. No samples were collected in Week 30 due the lack of fishing effort (Table 5).

The proportion estimates were used to estimate numbers of fish caught from each region for each fishery (Table 8). Since there were no genetic samples obtained from District 101 GNF weeks 36-39 (Table 4) and no samples for week 30 in the District 104 PSF (Table 5); those weeks were not represented in the regional estimates in Table 8. Table 8 also shows the estimated number of fish caught per region prior to Statistical Week 31. The Pacific Salmon Treaty allows for the harvest of a fixed percentage of Nass (for District 101) and Nass/Skeena (for District 104) sockeye prior to week 31.

## **DISCUSSION**

Chapter 2 of the 1999 Pacific Salmon Treaty specifies U.S. and Canada harvest sharing arrangements of Nass and Skeena River sockeye salmon in Northern Boundary fisheries. In Alaska's District 101 and District 104 sockeye fisheries, the United States is allowed to harvest a fixed percentage of the annual allowable harvest (AAH) of Nass and Skeena River sockeye salmon. Estimates of the stock-specific catch in these commercial fisheries were being provided by ADF&G using scale pattern analysis (SPA). This technique was replaced by genetic analysis in 2012.

Genetic markers are more stable than scale patterns and are not normally influenced by small environmental changes in short periods of time. Allelic frequency differences of genetic markers can be used to distinguish individual stocks of fish. These allele frequency differences can be reflective of adaptive measures taken by unique

stocks of fish to thrive in different environmental conditions, although these changes can often take many generations. Genetic stock identification is a powerful technique that takes advantage of these genetic differences to discriminate stocks of fish caught in a mixed stock fishery.

Auke Bay Laboratories has completed its genetic analysis of sockeye salmon caught in Districts 101 gillnet and District 104 purse seine fisheries for 2017. It should be recognized that while a total of 48 SNPs are currently used in the Southeast Alaska-British Columbia baseline, not all SNPs are likely to be equally informative.

## **CONCLUSION**

Our results indicate that a majority of sockeye salmon caught in the ADF&G District 101 GNF and District 104 PSF originated from Canadian stocks in 2017. Our results are in general agreement with the mark-recapture studies completed in the early 1980's (Pella et al., 1993), SPA completed since 1982 (Marshall, 1984), allozyme/freshwater age/parasitism analyses completed in the late 1980's (Pella et al., 1998), and SNP based genetic stock composition analyses completed since 2004. These correlations strongly suggest that all stock assessment methods have produced accurate and meaningful results in the management of these Northern Boundary fisheries. Compared with other methods, SNP genotyping is the most efficient method for stock assessment since it can be partially automated and the baseline does not require annual resampling. These advantages make it possible to use SNP markers to determine stock composition in a quicker time interval, allowing for improved management of the Northern Boundary fisheries. The similarity between stock composition estimates produced using scale pattern analysis and genetic analysis helps validate both approaches for determining stock assessments (Oliver 2009, Guthrie et al. 2009).

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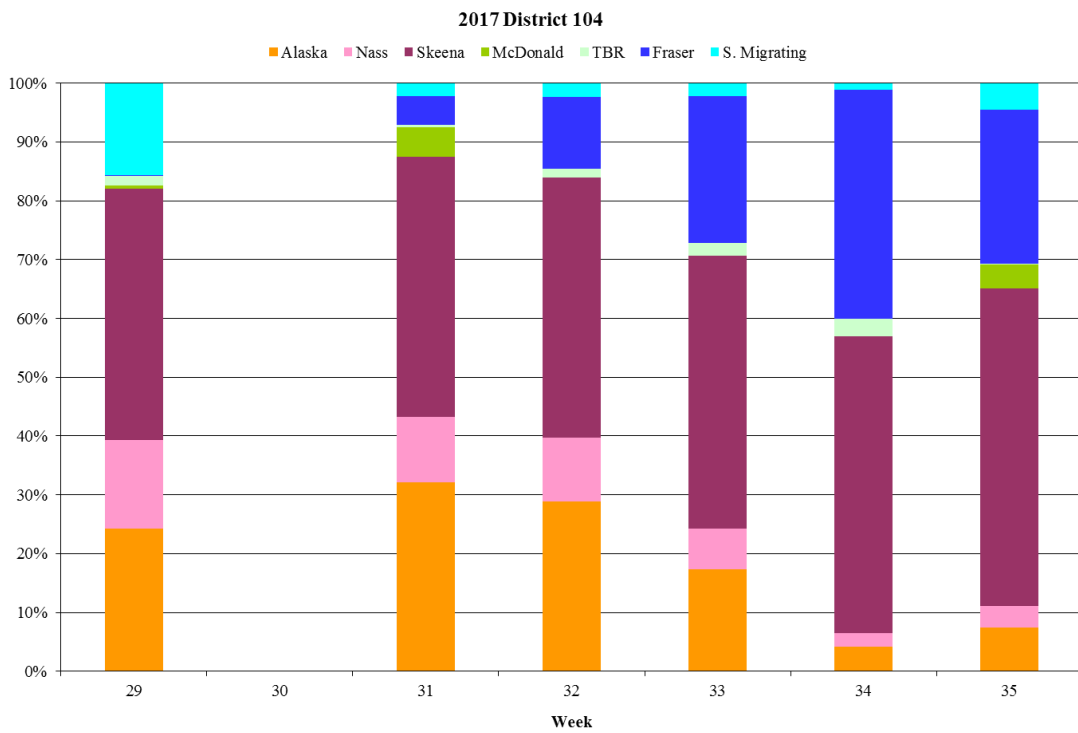
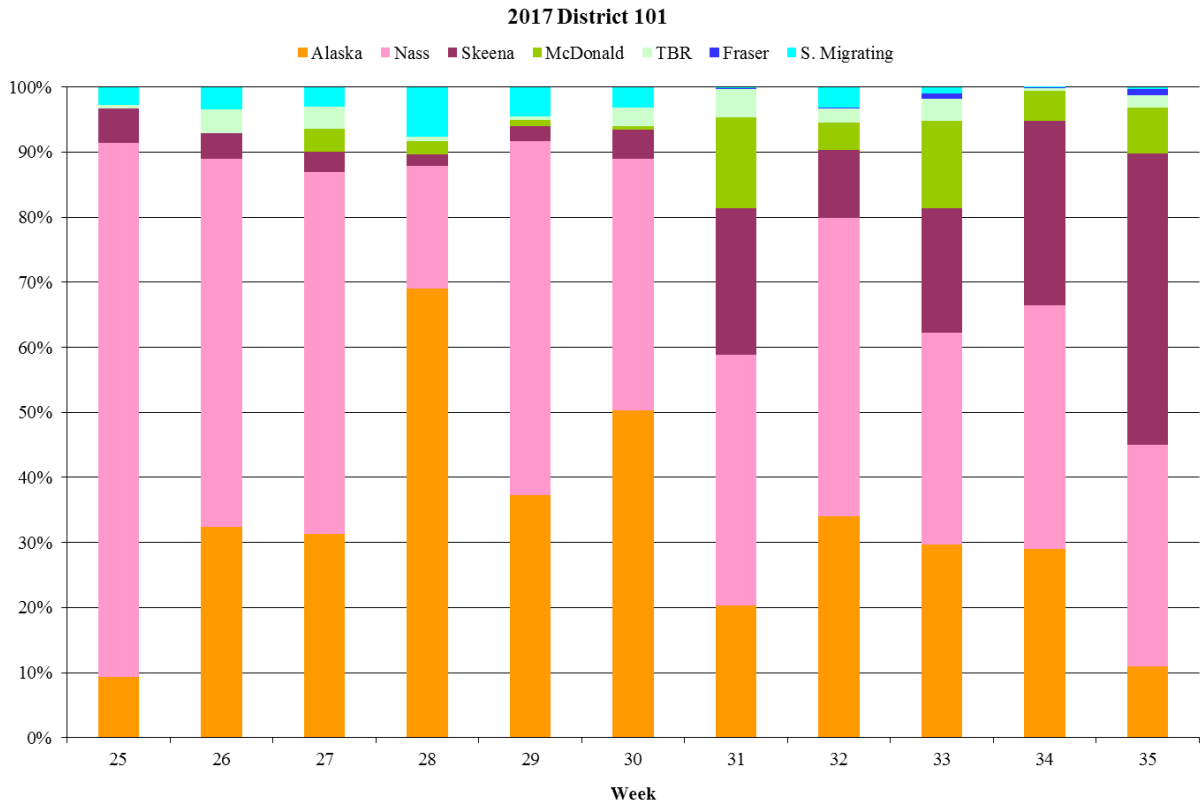
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**Figure 2.** 2017 sockeye stock group proportions for each statistical week from the ADF&G District 101 gillnet (top panel) and 104 purse seine fisheries (lower panel).

Table 6. Stock composition of weekly mixtures of sockeye salmon the 2017 District 101 commercial gillnet fishery.

|                      | Week 25 |      |             | Week 26 |      |             | Week 27 |      |             | Week 28 |      |             |
|----------------------|---------|------|-------------|---------|------|-------------|---------|------|-------------|---------|------|-------------|
|                      | Mean    | SD   | 95% CI      | Mean    | SD   | 95% CI      | Mean    | SD   | 95% CI      | Mean    | SD   | 95% CI      |
| Alaska               | 9.3     | 2.03 | (5.7,13.7)  | 32.4    | 3.17 | (26.4,38.8) | 31.4    | 4.48 | (22.7,39.9) | 69.0    | 4.81 | (57.1,76.3) |
| Nass River           | 82.2    | 2.56 | (76.8,86.9) | 56.5    | 3.30 | (50.0,62.9) | 55.6    | 3.86 | (47.8,62.9) | 18.9    | 2.58 | (14.1,24.2) |
| Skeena River         | 5.3     | 1.61 | (2.5,8.8)   | 4.0     | 1.36 | (1.7,7.0)   | 3.1     | 2.09 | (0.2,8.5)   | 1.8     | 1.08 | (0.0,4.3)   |
| McDonald Lake        | 0.0     | 0.18 | (0.0,0.0)   | 0.0     | 0.11 | (0.0,0.0)   | 3.5     | 3.63 | (0.0,11.3)  | 2.0     | 3.86 | (0.0,13.0)  |
| Transboundary Rivers | 0.5     | 0.89 | (0.0,3.2)   | 3.7     | 1.67 | (0.9,7.4)   | 3.4     | 1.65 | (0.8,7.1)   | 0.7     | 0.66 | (0.0,2.4)   |
| Fraser River         | 0.0     | 0.12 | (0.0,0.4)   | 0.1     | 0.16 | (0.0,0.5)   | 0.0     | 0.14 | (0.0,0.4)   | 0.0     | 0.12 | (0.0,0.3)   |
| South Migrating      | 2.7     | 1.20 | (0.8,5.5)   | 3.4     | 1.31 | (1.3,6.3)   | 3.0     | 1.35 | (0.9,6.1)   | 7.6     | 1.86 | (4.3,11.6)  |

|                      | Week 29 |      |             | Week 30 |      |             | Week 31 |      |             | Week 32 |      |             |
|----------------------|---------|------|-------------|---------|------|-------------|---------|------|-------------|---------|------|-------------|
|                      | Mean    | SD   | 95% CI      | Mean    | SD   | 95% CI      | Mean    | SD   | 95% CI      | Mean    | SD   | 95% CI      |
| Alaska               | 37.3    | 3.67 | (29.4,44.1) | 50.3    | 3.68 | (42.4,57.1) | 20.4    | 3.44 | (14.0,27.4) | 34.0    | 6.86 | (20.0,44.4) |
| Nass River           | 54.4    | 3.19 | (48.2,60.6) | 38.7    | 3.20 | (32.5,45.1) | 38.4    | 3.61 | (31.4,45.5) | 45.9    | 3.26 | (39.5,52.3) |
| Skeena River         | 2.3     | 1.20 | (0.5,5.1)   | 4.6     | 1.61 | (1.9,8.2)   | 22.6    | 3.35 | (16.5,29.6) | 10.5    | 2.14 | (6.7,15.1)  |
| McDonald Lake        | 0.9     | 2.05 | (0.0,7.3)   | 0.4     | 1.71 | (0.0,6.8)   | 14.0    | 2.99 | (8.2,20.1)  | 4.2     | 5.99 | (0.0,17.2)  |
| Transboundary Rivers | 0.6     | 0.58 | (0.0,2.2)   | 2.9     | 1.68 | (0.6,7.0)   | 4.4     | 2.01 | (0.6,8.7)   | 2.2     | 1.68 | (0.0,6.0)   |
| Fraser River         | 0.1     | 0.22 | (0.0,0.7)   | 0.0     | 0.11 | (0.0,0.3)   | 0.0     | 0.12 | (0.0,0.4)   | 0.0     | 0.12 | (0.0,0.3)   |
| South Migrating      | 4.5     | 1.49 | (2.0,7.7)   | 3.1     | 1.58 | (0.0,6.6)   | 0.2     | 0.53 | (0.0,1.8)   | 3.2     | 1.59 | (0.5,6.7)   |

|                      | Week 33 |      |             | Week 34 |      |             | Week 35 |      |             |
|----------------------|---------|------|-------------|---------|------|-------------|---------|------|-------------|
|                      | Mean    | SD   | 95% CI      | Mean    | SD   | 95% CI      | Mean    | SD   | 95% CI      |
| Alaska               | 29.7    | 3.99 | (22.2,37.8) | 29.0    | 5.80 | (18.2,40.4) | 10.9    | 3.58 | (5.0,19.2)  |
| Nass River           | 32.6    | 3.43 | (25.9,39.4) | 37.4    | 4.94 | (28.0,47.3) | 34.2    | 3.58 | (27.3,41.3) |
| Skeena River         | 19.1    | 2.98 | (13.7,25.4) | 28.4    | 4.58 | (19.9,37.9) | 44.7    | 3.74 | (37.5,52.1) |
| McDonald Lake        | 13.4    | 3.34 | (7.1,20.1)  | 4.7     | 4.67 | (0.0,14.8)  | 7.0     | 3.30 | (0.0,13.5)  |
| Transboundary Rivers | 3.4     | 2.20 | (0.0,8.3)   | 0.4     | 0.80 | (0.0,2.8)   | 2.0     | 1.85 | (0.0,6.3)   |
| Fraser River         | 0.8     | 0.65 | (0.0,2.4)   | 0.1     | 0.24 | (0.0,0.7)   | 0.9     | 0.71 | (0.1,2.7)   |
| South Migrating      | 1.0     | 1.17 | (0.0,3.8)   | 0.1     | 0.26 | (0.0,0.7)   | 0.3     | 0.76 | (0.0,2.8)   |

Table 7. Stock composition of weekly mixtures of sockeye salmon in the 2017 District 104 commercial purse seine fishery.

|                      | Week 29 |      |             | Week 31 |      |             | Week 32 |      |             |
|----------------------|---------|------|-------------|---------|------|-------------|---------|------|-------------|
|                      | Mean    | SD   | 95% CI      | Mean    | SD   | 95% CI      | Mean    | SD   | 95% CI      |
| Alaska               | 24.3    | 3.25 | (18.0,30.7) | 32.2    | 3.23 | (26.0,38.7) | 28.9    | 3.20 | (22.8,35.3) |
| Nass River           | 15.1    | 2.63 | (10.3,20.6) | 11.1    | 2.01 | (7.4,15.2)  | 10.9    | 2.13 | (7.0,15.3)  |
| Skeena River         | 42.8    | 3.42 | (36.2,49.6) | 44.3    | 2.98 | (38.5,50.2) | 44.2    | 3.22 | (38.0,50.5) |
| McDonald Lake        | 0.5     | 1.38 | (0.0,5.1)   | 4.9     | 1.86 | (1.4,8.8)   | 0.1     | 0.48 | (0.0,0.9)   |
| Transboundary Rivers | 1.6     | 1.58 | (0.0,5.6)   | 0.4     | 0.74 | (0.0,2.6)   | 1.4     | 1.52 | (0.0,5.1)   |
| Fraser River         | 0.0     | 0.13 | (0.0,0.4)   | 4.9     | 1.24 | (2.8,7.6)   | 12.2    | 2.03 | (8.5,16.5)  |
| South Migrating      | 15.7    | 2.58 | (10.9,20.9) | 2.1     | 1.29 | (0.1,5.0)   | 2.4     | 1.22 | (0.6,5.2)   |

|                      | Week 33 |      |             | Week 34 |      |             | Week 35 |      |             |
|----------------------|---------|------|-------------|---------|------|-------------|---------|------|-------------|
|                      | Mean    | SD   | 95% CI      | Mean    | SD   | 95% CI      | Mean    | SD   | 95% CI      |
| Alaska               | 17.3    | 2.54 | (12.6,22.5) | 4.2     | 1.38 | (1.9,7.2)   | 7.4     | 2.44 | (3.4,13.0)  |
| Nass River           | 6.9     | 1.90 | (3.5,11.0)  | 2.3     | 0.87 | (0.9,4.3)   | 3.6     | 1.25 | (1.6,6.4)   |
| Skeena River         | 46.4    | 3.08 | (40.4,52.5) | 50.5    | 3.04 | (44.5,56.4) | 54.1    | 3.19 | (47.9,60.3) |
| McDonald Lake        | 0.0     | 0.28 | (0.0,0.3)   | 0.0     | 0.25 | (0.0,0.3)   | 4.0     | 1.91 | (0.0,7.8)   |
| Transboundary Rivers | 2.2     | 1.62 | (0.0,5.7)   | 3.0     | 1.55 | (0.2,6.4)   | 0.2     | 0.37 | (0.0,1.3)   |
| Fraser River         | 24.9    | 2.52 | (20.1,30.0) | 38.9    | 2.86 | (33.4,44.5) | 26.2    | 2.79 | (20.9,31.8) |
| South Migrating      | 2.2     | 1.23 | (0.3,5.0)   | 1.1     | 0.76 | (0.1,3.0)   | 4.5     | 1.75 | (1.4,8.3)   |

**Table 8.** Estimated numbers of sockeye salmon caught in the 2017 District 101 gillnet and 104 seine fisheries prior to statistical week 31 and throughout all statistical weeks analyzed (see Tables 4&5). The estimate for district 104 does not include statistical week 30 due to small sample size.

| <i>Region</i> | <i>Area</i>          | <i>District 101 Gillnet</i> |              | <i>District 104 Seine</i> |              |
|---------------|----------------------|-----------------------------|--------------|---------------------------|--------------|
|               |                      | <i>Prior to 31</i>          | <i>Total</i> | <i>Prior to 31</i>        | <i>Total</i> |
| 1             | Alaska               | 4,311                       | 7,474        | 1,817                     | 16,680       |
| 2             | Nass River           | 8,141                       | 12,344       | 1,129                     | 6,869        |
| 3             | Skeena River         | 502                         | 2,557        | 3,207                     | 43,926       |
| 4             | McDonald Lake        | 170                         | 1,002        | 40                        | 1,790        |
| 5             | Transboundary Rivers | 210                         | 520          | 123                       | 1,273        |
| 6             | Fraser River         | 6                           | 26           | 3                         | 18,165       |
| 7             | South Migrating      | 546                         | 704          | 1,173                     | 3,275        |
| <i>Totals</i> |                      | 13,886                      | 24,628       | 7,492                     | 91,977       |