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Estimation of abundance and run timing for coho salmon originating from the upper Kuskokwim River from Bethel test fishery samples

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ABSTRACT:

Coho salmon spawning in the upper portion of the Kuskokwim River upstream of the Takotna River represent an important component of the genetic and life history variation for this species in the watershed. Mark-recapture studies conducted in the Kuskokwim River suggest that coho salmon spawning in tributaries at increasing distances from the river mouth have progressively earlier run timing. Early run timing can pose a management concern because information to assess run abundance is the least reliable early in the fishing season resulting in a greater risk of overharvesting early-run stocks. In this study, coho salmon were sampled from the Bethel test fishery, an annual test fishery conducted in the lower Kuskokwim River near Bethel, in 2008-2010. Mixed-stock analysis using genetic characters was used to estimate the contributions of three stock groups to the test fishery samples (Upper Kuskokwim River, coho salmon spawning in tributaries upstream of the Takotna River; Lower Kuskokwim River, coho salmon spawning in tributaries from the river mouth to the Takotna River; and Kuskokwim Bay, coho salmon spawning in the Middle Fork Goodnews, Arolik, and Kanektok rivers) to determine their relative abundance and the run timing of Upper Kuskokwim River coho salmon over a three-year period. Annual average stock contributions were consistent over the three years of the study and ranged from 5-6% for Upper Kuskokwim River, 88-92% for Lower Kuskokwim River, and 3-5% for Kuskokwim Bay. The test fishery was divided into four time strata corresponding to the quartiles of the fishery in each year to investigate run timing of Upper Kuskokwim River. Upper Kuskokwim River was present in each quartile of the fishery in each sampling year, with the numerically largest estimates occurring in the 4th quartile. Stock composition estimates for the first 50% of the fishery for Upper Kuskokwim River were not larger than estimates for the second 50%, and did not provide evidence for early run timing for this stock.

Key words: Bethel test fishery, coho salmon, Kuskokwim River, mixed-stock analysis, run timing

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INTRODUCTION:

Populations of salmon spawning within a watershed can demonstrate population-specific patterns of timing of entry into freshwater (e.g., Hess and Narum 2011; Flannery et al. 2010; Eiler et al. 2004). Timing of freshwater entry is under partial genetic control to ensure that spawning occurs at the appropriate date and is influenced by a variety of factors, including migration length and water conditions encountered during migration and at the spawning site (reviewed in Quinn 2005). Though timing of freshwater entry can be difficult to predict (Quinn 2005), knowledge of timing of entry is an important component of fishery management in lower river fisheries that harvest mixed stocks in order to allow for adequate harvest while conserving genetic and life history diversity (e.g., Vähä et al 2010, Keefer et al. 2004, Shaklee et al. 1999).

The Kuskokwim River (Figure 1) is the second largest river in Alaska, draining an area of 150,000 km^2 (Linderman and Bergstrom 2009). It originates from the northwestern side of the Alaska Range, and flows approximately 1,500 km where it drains into Kuskokwim Bay and the Bering Sea. Coho salmon *Oncorhynchus kisutch* are the second most abundant Pacific salmon in the Kuskokwim River, and are a mainstay to the ecology and economy of the region (Whitmore et al. 2005). Coho salmon have the widest spawning distribution in the watershed, and are the only Pacific salmon to thrive in the very upper reaches of the Kuskokwim River (Stokes 1985). The Kuskokwim River supports the largest commercial coho salmon fishery in western Alaska; the 2001-2010 ten-year average harvest was 319, 567 fish (Brazil et al. 2011). Revenue generated from coho salmon harvests contributes greatly to the economy in the region, where employment opportunities are generally low (Linderman and Bergstrom 2009).

Coho salmon return to the Kuskokwim River from early July through fall to spawn in tributaries

ranging from the river mouth to the South, East, and North Forks in the upper watershed (Figure 1). Commercial fisheries occur in the lower Kuskokwim River (District W1; Figure 1). Management decisions to open and close the fishery are based on daily abundance indices from a gillnet test fishery operated near Bethel; harvest trends in the commercial and subsistence fisheries; and from tributary escapements monitored using weirs in the Kwethluk (rkm 216), Tuluksak (rkm 248), George (rkm 453), Tatlawiksuk (rkm 568), Kogrukluk (rkm 710), and Takotna (rkm 835) rivers (Molyneaux and Brannian 2006) (Figure1).

Coho salmon spawning in upper Kuskokwim River tributaries represent an important component of the life history and genetic variation contained in the watershed. Coho salmon typically do not migrate more than approximately 250 km upstream in large rivers to spawn (Sandercock 1991), but spawning migrations in the Kuskokwim River extend 1,000 km upstream from the river mouth, and are among the longest documented for this species (Whitmore et al. 2005; Sandercock 1991). Coho salmon in the Kuskokwim River are subdivided into two genetic diversity groups, one comprising spawners in lower Kuskokwim River tributaries upstream to the Takotna River, and one comprising spawners in the upper Kuskowkim River (Olsen et al. 2011, Crane et al. 2007).

Coho salmon tagged in the Kuskokwim River near Kalskag (rkm 263, Figure 1) and recaptured at weirs on the George, Tatlawiksuk, Kogrukluk, and Takotna rivers show progressively earlier passage timing at Kalskag for fish migrating greater distances from the river mouth (Schaberg et al. 2010). Though subsistence fishers in Nikolai and Telida report the presence of coho salmon in the South Fork Kuskokwim River, Salmon River, and Highpower Creek in August through October (Stokes 1985), there is no specific run timing information for coho salmon upstream of

the Takotna River.

Given the existing patterns of migration for currently monitored coho salmon stocks, we hypothesized that coho salmon spawning in the upper Kuskokwim River may have earlier freshwater entry timing (run timing) than lower river stocks. Stocks of salmon with early run timing are a concern because information to assess run abundance is the least reliable early in the fishing season resulting in a greater risk overharvesting these stocks (Gilk and Molyneaux 2004). Further, if overharvest does occur in upriver stocks, there are no resources to assess the impacts on escapement upstream of the Takotna River.

Mixed-stock analysis (MSA, Utter and Ryman 1993, Pella and Milner 1987) uses genetic characters to estimate the stock components of mixtures given the underlying frequency of genetic characters in stocks contributing to the mixture ("baseline"). It has been successfully used to estimate stock-specific run timing of Pacific salmon showing similar amounts of genetic variation as that detected in coho salmon in the Kuskokwim River. Examples include Chinook salmon *O. tshawytscha* in the Yukon River (Templin et al. 2005), Fraser River (Beacham et al. 2003), and Columbia River (Shaklee et al. 1999); sockeye salmon *O. nerka* in the Nass (Beacham and Wood 1999), and Skeena rivers (Beacham et al. 2000); and chum salmon *O. keta* in the Yukon River (e.g., Flannery et al. 2010). In this project, we determined the relative proportion of three stock groups of Kuskokwim River area coho salmon to fish sampled in a test fishery conducted from June to late August in the lower Kuskokwim River near Bethel in 2008, 2009, and 2010, with the overall goal of determining the relative abundance and run timing of coho salmon returning to spawn in the upper Kuskokwim River.

OBJECTIVES:

Objective 1. Determine the relative contribution of three stocks of coho salmon to fish sampled from the Bethel test fishery.

Contribution estimates were made for three stock groups to coho salmon sampled from the Bethel test fishery 2008-2010: Upper Kuskokwim River (coho salmon spawning in the Kuskokwim River tributaries upstream of the Takotna River); Lower Kuskokwim River (coho salmon spawning in tributaries of the Kuskokwim River from the river mouth to the Takotna River), and Kuskokwim Bay (Kanektok River, Arolik River, and Goodnews River). Upper Kuskokwim River made small contributions to the Bethel test fishery. During the three years of this study, annual contribution estimates ranged from 5-6%. Zero was not in the 95% credibility intervals for individual stock contribution estimates, indicating that the genetic distinctiveness of upper Kuskokwim River coho salmon and the mixture sample sizes used for the analysis were adequate to successfully detect small contributions to the fishery for this stock. Lower Kuskokwim River was the largest contributor to Bethel test fishery samples, with estimates ranging from 88-92%, while estimates for Kuskokwim Bay ranged from 3-5%.

Objective 2. Determine if run timing of coho salmon at the Bethel test fishery is stock-specific. The mixture samples were divided into four time strata corresponding to the four quartiles of the Bethel test fishery in each sampling year to investigate run timing for Upper Kuskokwim River. In each sampling year, Upper Kuskokwim River was present in all four quartiles of the Bethel test fishery, with the numerically largest estimates occurring in the 4th quartile. We did not find evidence of early run timing for this stock; instead, at least in 2008, larger contribution estimates were observed during the latter part of the fishery. Though mixture sample sizes were large

enough to detect the presence of Upper Kuskokwim River in the fishery samples, mixture sample sizes were not large enough to detect small changes in stock composition estimates with certainty for each year of the study.   Individual based-analyses were also used to evaluate run timing of upper Kuskokwim River coho salmon. Cumulative catch curves for upper Kuskokwim River coho salmon were developed, and suggest that this stock had a slightly later timing of river entry in 2008 and 2009 than the Kuskowkim Area stock as a whole.

Objective 3. Determine if relative contributions of the stocks of coho salmon vary among years. The contribution estimates for three stocks of coho salmon (Upper Kuskokwim River, Lower Kuskokwim River, and Kuskokwim Bay) were consistent across all three years of the study.

METHODS:

Study area

The Bethel drift gillnet test fishery (BTF) is conducted approximately 5km upstream of Bethel at about the midpoint of commercial fishing district W1 of the lower Kuskokwim River (Molyneaux 2003) (Figure 1). The BTF has been operated annually since 1984 and provides a relative index for salmon migratory timing and run strength (Molyneaux 2003). Test fishing occurs daily from early June to late August. The test fishery entails a series of three successive drifts using 5-3/8" mesh gillnets beginning one hour after slack high tide (Brazil et al. 2011). The project is conducted by Alaska Department of Fish and Game (ADFG), with assistance from Orutsararmiut Native Council (ONC).

Sample collection

Fin clips from up to 50 coho salmon per day were taken from coho salmon collected in the BTF by ADFG and ONC, from the first day coho salmon were caught until the last day of test fishing. Fin clips were preserved in a 250ml bottle in 95% ethanol labeled with the day's date, and shipped to the Conservation Genetics Laboratory, U.S. Fish and Wildlife Service, for long term storage and laboratory analysis. Each year, samples were stratified post-season into temporal strata, one stratum for each quartile of the BTF. Quartiles were determined from the cumulative daily catch per unit effort (CPUE). Stock composition estimates were made for each quartile by randomly sampling 300 fish proportional to the day's CPUE within each quartile.

Coho salmon were also sampled from District W1 commercial fishery openings in all three sampling years (Table 1). These samples were not used in the data analysis because the BTF samples provided adequate representation of the run.

Laboratory analysis

Total genomic DNA was extracted from approximately 25mg of fin tissue from each randomly

sampled fish with Dneasy™ DNA isolation kits (Qiagen Inc., Valencia, CA). Genetic variation

was assayed at fourteen microsatellite loci: Ocl8 (Condrey and Bentzen 1998; Oke 2, 3, and 4

(Buchholz et al. 1999); Oki1, 2, and 11 (Smith et al. 1998); Omy1011 (Morris et al 1996); Oneu3

(Scribner et al. 1996); Ots2M (Grieg and Banks 1999); Ots101 (Small et al. 1998); Ots105

(Nelson 1998); Ots213 (Grieg et al. 2003); and Ssa407 (Cairney et al. 2000). Polymerase chain

reaction (PCR) amplifications of microsatellite loci were carried out in 96-well plates in 10ul

reaction volumes using 30-50ng DNA, 1.5-2 mM $MgCl_2$, 0.8-1mM dNTPs, 0.1-0.4uM labeled

forward primer, and 0.4uM unlabeled reverse primer using a BioRad thermocycler. Cycling

conditions were 1 cycle of 2 min at 92°; 30 cycles of 15 sec at 92°, 15 sec at 56°-58°, and 30 sec

at 72°; with a final extension for 10 min at 72°. The PCR products were size fractioned using an

Applied Biosystems 3730 Genetic Analyzer. Applied Biosystems GeneScan™-600 LIZ® size

standard, 20-600 bases, was loaded in all lanes for internal lane standards. Two researchers

scored alleles independently. Samples with score discrepancies between researchers were re-

amplified at the loci in question and rescored to resolve score discrepancies. Lastly, PCR

amplifications for all loci were conducted a second time for 8% of the samples. The PCR

products were size fractioned and scored to check and correct for laboratory errors.

Data analysis

Evaluation of baseline for mixture analysis

Baseline data used for estimating the stock composition of the BTF samples are those described

in Olsen et al (2011) and Crane et al. (2007) (Table 1, Figure 1), with the following

modifications. First, data described in Olsen et al. (2011) and Crane et al. (2007) were obtained

using a Licor sequencing platform. Allele sizes were standardized to the ABI 3730. Archive

samples (Middle Fork Goodnews River 2001, Arolik River 1997, Kanektok River 2001, Kisaralik River 1997, George River 2001, Kogrukluk River 2001, Tatlawiksuk River 2001, and Takotna River 2001) were analyzed using the ABI 3730 to ensure consistency of alleles for data collected between the two platforms. Second, additional baseline samples were added to the baseline from Eek River, Salmon (Aniak) River, Stony River, Swift River, Big River, Middle Fork Windy Fork, South Fork, and Tin Creek (see Table 1).

Exact tests conducted in Genepop version 4 (Rousset 2008; Raymond and Rousset 1995) were used to test for variation in allele frequencies in samples collected at the same tributary in different years ("temporal samples"). In addition, analysis of molecular variance (AMOVA, Excoffier et al. 1992) was used to evaluate the relative proportion of genetic variation due to temporal differences within population samples and variation among population samples using Arlequin version 3.11 (Excoffier et al. 2005).

Temporal samples were pooled into a single population sample to reduce error in population allele frequency estimates due to drift in allele frequencies between sampling years (Waples 1990). Conformance of genotypic frequencies to Hardy-Weinberg expectation was evaluated using exact tests in Genepop. Critical values (tablewide $\alpha=0.05$) were adjusted for multiple tests for loci within populations (14 tests) and each locus across populations (21 tests) using a sequential Bonferroni procedure (Rice 1989). Linkage disequilibrium was also evaluated using exact tests between all pairs of loci within all populations. If significant disequilibrium existed between a pair of loci in 50% or more of the populations, one of the loci was removed for the remaining analyses (McGlauflin et al. 2011). Cavalli-Sforza and Edwards (1967) chord distances were calculated from allele frequencies for all pairwise combinations of populations in

Phylip version 3.68 (Felsenstein 2004). Distances were used in a multidimensional scaling

analysis conducted in NTSYS version 2.10p (Exeter Software, NJ) to visualize spatial

relationships among populations.

Using conditional maximum likelihood of simulated mixtures, Crane et al. (2007) found that

three stock groups can be identified in mixtures of coho salmon from the Kuskokwim Area to

approximately 90% of the true value of 100%: Upper Kuskokwim River (South Fork

Kuskokwim River, Highpower Creek), Lower Kuskokwim River (Kwethluk to Takotna River),

and Kuskokwim Bay (Middle Fork Goodnews, Arolik, and Kanektok rivers).

We evaluated the performance of the updated baseline and the three reporting groups defined

above using three methods. First, for each reporting group, 1000 simulated mixtures of 300 fish

were generated from the baseline allele frequencies using Hardy-Weinberg proportions, with

each baseline population within each reporting group contributing equally. Perfect performance

would result in mean stock contribution estimates of approximately 100% for a given reporting

group. Mixtures were created and stock compositions were estimated using Oncor (available at

http://www.montana.edu/kalinowski/Software/ONCOR.htm; Anderson et al. 2008).

Second, we estimated the stock composition of four mixtures of known origin because analysis

of simulated mixtures can overestimate the accuracy and precision of stock composition

estimates (Anderson et al. 2008, Koljonen et al. 2005). Mixtures were created by randomly

sampling 200 or 300 multilocus genotypes from the baseline, deleting mixture genotypes from

the baseline and recompiling baseline allele frequencies. The known mixture proportions were:

Mixture 1: 100% Upper Kuskokwim (N=200); Mixture 2: 100% Lower Kuskokwim (N=200);

Mixture 3: 100% Kuskokwim Bay (N=200); and Mixture 4: 2% upper Kuskokwim River, 91% Lower Kuskokwim River, and 7% Kuskokwim Bay (N=300), following stock proportions estimated from the District W1 commercial fishery sample described in Crane et al. (2007).

The stock composition of each of the mixtures was estimated using Bayesian mixture modeling (Pella and Masuda 2001) as implemented in Bayes (ftp://wwwabl.afs.noaa.gov/sida/mixture-analysis/bayes). Initial starting proportions for three chains of 50,000 interations each were 95% from each reporting group, with all populations contributing equally, and 5% distributed evenly among the remaining populations. Values for flat genetic prior parameters were determined as described in Pella and Masuda (2001). The Raftery and Lewis (1996) diagnostic was used to verify that the number of iterations was sufficient. Convergence among chains was determined using Gelman and Rubin (1992) shrink factors; shrink factors of less than 1.2 were used to indicate convergence. The mean, standard deviation, and 95% credibility intervals of sample stock composition estimates were generated after a burn-in discard of the first half of the samples.

Lastly, we evaluated the baseline for its use in assignment of individual fish from a mixture to reporting group. Assignment tests were conducted in Oncor using the "leave-one-out" option, where each multilocus genotype is removed from the baseline, and assigned to its most probable stock of origin. In the known mixture tests, mixture individuals were assigned to their reporting group of origin based on the greatest average posterior source probability (Pella and Masuda 2001, Koljonen et al. 2005).

Mixture analysis

Since contribution estimates for Upper Kuskokwim River are likely to be small, stock compositions for each BTF quartile in each year were estimated using Bayesian mixture modeling with the program Bayes (Pella and Masuda 2001). This method produces estimates that are less biased when stock contributions are extreme (Pella and Masuda 2001; Reynolds and Templin 2004). Individuals missing data for four or more loci or deemed not to be coho salmon were deleted from the mixture prior to analysis. Estimates for strata within years were combined into a single estimate per year by weighting them according to the cumulative CPUE within each quartile following Flannery et al. (2010).

We evaluated stock-specific run timing in two ways. First, we calculated the proportion of times the Upper Kuskokwim River estimate for Quartile 1 and Quartile 2 exceeded the estimate for both Quartile 3 and Quartile 4 from the 75,000 posterior Markov Chain Monte Carlo stock estimates to determine the probability that the Upper Kuskokwim River had an earlier run timing. Second, posterior probabilities were used to assign individuals to stock of origin based on the greatest average posterior source probability (Upper Kuskokwim River and Lower Kuskokwim River/Kuskokwim Bay) and cumulative catch curves were developed for the Upper Kuskokwim River stock as an index of run timing.

RESULTS:

Sample collection

In 2008, 2,562 coho salmon were sampled from the BTF. The first coho salmon appeared in the BTF on 11 July, and test fishery sampling continued daily until 11 September (Table 1). In 2009, 1,846 coho salmon were sampled, with the first coho salmon appearing in the BTF on 11 July, and sampling continued until the BTF end date of 24 August. During the first two weeks of

the 2009 BTF, all fish were placed into a single 200ml bottle, rather than individual bottles labeled with the day's date. In 2010, the coho salmon return was weak (Brazil et al. 2011); 846 coho salmon were sampled from the BTF between 16 July and 24 August.

In 2008 and 2009, enough coho salmon were sampled to randomly sample 300 fish per quartile, proportional to the daily CPUE (Figure 2). Though the test fishery was operational until 11 September in 2008, we used 24 August as the end date, for consistency with the other years of the study. All fish were used for mixture analysis in 2010 because so few fish were caught; however, the samples analyzed per day were representative of the daily CPUE (Figure 2).

Evaluation of baseline for mixture analysis
Allele frequency data for baseline populations are shown in Appendix 1. Significant deviation of genotypic frequencies from Hardy-Weinberg expectation was detected in 28 of 283 tests. Five of these tests were judged significant after adjusting for multiple tests for each locus; *Ots101* in Kanektok and Arolik rivers; *Ssa407* in Salmon River and Tin Creek, and *Ots213* in Takotna River (Appendix 1).   No population had more than one locus not conforming to Hardy-Weinberg expectation after adjusting for 14 multiple tests within each population (Appendix 1). No pair of loci were found to be in linkage disequilibrium ($P<0.05$) in greater than 50% of the populations; all loci were retained for subsequent analyses.

Heterogeneity was tested for temporal samples taken in Highpower Creek, and the Takotna, Tatlawiksuk, Swift, Kogrukluk, George, Salmon, Kisaralik, Kanektok, and Middle Fork Goodnews rivers. Of these, significant differences in allele frequencies were detected in the Kisaralik ($P=0.0039$), Takotna ($P<0.001$), and Swift rivers ($P<0.001$) after adjustment for

multiple tests. Analysis of molecular variance (AMOVA) of all population samples found that 0.31% of the genetic variation in the coho salmon collections was due to variation among temporal samples, and 2.38% was due to genetic variation among populations. Both components of variation were significantly greater than zero.   In the multidimensional scaling analysis, populations from the upper Kuskokwim River (numbers 15-21) were relatively dispersed, and clearly separated from populations from Kuskokwim Bay (numbers 1-3) and from the Kuskokwim River mouth to the Takotna River (numbers 4-14) (Figure 3).

The mean mixture composition estimates generated by Oncor were 0.99 (95% CI 0.98-1.0) for Upper Kuskokwim River, 0.96 (95% CI 0.90-1.0) for Lower Kuskokwim River, and 0.88 for Kuskokwim Bay (95% CI 0.80-0.96). Virtually no misallocation (<0.01) occurred to Upper Kuskokwim River in the simulations for Lower Kuskokwim and Kuskokwim Bay reporting groups.

For the known mixture tests, all three mixtures composed 100% from a single reporting group had stock contribution estimates exceeding 0.90 (Table 2). Similar to the simulation analysis, misallocation to Upper Kuskokwim River in the analysis of the 100% Lower Kuskokwim River and Kuskokwim Bay mixtures was negligible, less than 0.01. The mixture estimate for Upper Kuskokwim River for the realistic fishery (Mixture 4) was 0.022, very close to its expectation (0.02). Further, the 95% credibility interval did not contain zero, indicating that with mixture sample sizes of 300, mixed-stock analysis should be able to detect small contributions of this reporting group.

Assignment tests conducted in Oncor showed that 93% of the individuals from the Upper

Kuskokwim River were correctly assigned to Upper Kuskokwim, and that 96% of the individuals

from Lower Kuskokwim River and Kuskokwim Bay were correctly assigned. Posterior

probabilities assigned all fish in known-origin Mixture 1 to Upper Kuskokwim River, and

assigned no fish in Mixture 2 or 3 to Upper Kuskokwim River. In Mixture 4, the realistic fishery

test, 4 of 6 fish from Upper Kuskokwim were assigned to Upper Kuskokwim River, and no fish

from Lower Kuskokwim River and Kuskokwim Bay were missassigned to Upper Kuskokwim

River.

Mixture Analysis

Objective 1, annual contribution estimates:--From 2008-2010, the annual average contribution of

Upper Kuskokwim River coho salmon to the BTF was very consistent, ranging from 0.05 to

0.06. Zero was not included in the 95% confidence interval for any estimate, an indication that

Upper Kuskokwim River coho salmon are detectable in the fishery samples, even though the

estimates are small. The majority of fish were from Lower Kuskokwim River, with estimates

ranging from 0.88 to 0.92. Kuskokwim Bay was a small contributor, ranging from 0.03 to 0.05

percent. Zero was included in the 95% confidence interval for one of the three sampling years.

Objectives 2 and 3, patterns within and across years:--The stock contribution estimates were

consistent across quartiles and across years (Table 3, Figure 4). Contribution estimates for

Upper Kuskokwim River coho salmon ranged from 0.02 to 0.10 in 2008, from 0.03 to 0.08 in

2009, and from 0.04 to 0.11 in 2010. For each sampling year, the largest estimate for Upper

Kuskokwim River appeared in Quartile 4. Stock contribution estimates within years for Lower

Kuskokwim River varied little, ranging from 0.87 to 0.95 in 2008, 0.85 to 0.96 in 2009, and 0.80

to 0.95 in 2010. Stock contribution estimates did not vary for Kuskokwim Bay in 2008 and were

consistently around 0.03. Zero was in the 95% credibility interval for each quartile estimate. In

2008 and 2009, contribution estimates for Kuskokwim Bay ranged from 0.01 to 0.11 and from

0.002 to 0.14, respectively. Zero was not included in the 95% credibility interval for Quartile 3

in 2009 and 2010.

The posterior probability that the stock contribution estimate for the Upper Kuskokwim River in

the first 50% of the BTF exceeded the estimate for the second 50% of the BTF was 0 for 2008,

0.071 for 2009, and 0.053 for 2010. Cumulative catch curves for coho salmon in the BTF, the

BTF-Upper Kuskokwim River fish only, and the District W1 commercial fishery are shown in

Figure 5. Median passage dates for the Upper Kuskokwim coho stock were August 16 in 2008,

August 9 in 2009, and August 8 in 2010. Upper Kuskokwim River coho salmon were present in

the BTF until the end of the fishery in all three years. The cumulative catch curves suggest that

Upper Kuskokwim River fish had a slightly later timing of river entry in 2008 and 2009 than the

Kuskowkim Area stock as a whole, but that in 2010 timing of river entry was relatively

compressed.

DISCUSSION:

Baseline

Olsen et al. (2011) found that coho salmon distributed from Kuskokwim Bay to Norton Sound

subdivide into two genetic diversity groups, one comprising inland-spawning salmon in the

Kuskokwim and Yukon Rivers, and one comprising salmon spawning in coastal areas. The

demarcation for the two groups in the Kuskokwim River was the confluence of the Takotna

River (rkm 752), with the inland group occurring upstream of the confluence and the coastal

group occurring downstream inclusive of the Takotna River. Olsen et al. (2011) speculated that historic effects (vicariance through stream capture events, or secondary contact among genetic lineages associated with different refugia during Pleistocene glaciations) could be responsible for this regional pattern. Alternatively, the longer and more complex freshwater migrations experienced by interior-spawning salmon may favor increased homing precision and thus reduce gene flow among coastal and interior groups (Olsen et al. 2011).

This study added data from seven additional collections to those reported in Olsen et al. (2011). The genetic distinction between the upper and lower river groups was maintained with the additional collections (Figure 3); average pairwise F_{ST} between upper and lower river collections was 0.067, but only 0.009 between lower river collections. Analysis of mixtures of known origin demonstrated that the genetic divergence between coho salmon spawning in the upper and lower Kuskokwim River permits accurate stock composition estimation and individual assignment.

Aside from the underlying genetic differentiation among the stock groups, other factors that may affect accuracy and precision of estimates of stock proportions and stock assignments are incomplete baseline, sampling variation in baseline stocks, and temporal changes in allele frequencies (Koljonen et al. 2005). All major spawning populations of coho salmon in tributaries draining into Kuskokwim Bay and the lower Kuskokwim River were represented in the Lower Kuskokwim River and Kuskokwim Bay stock groups. All major tributaries where coho salmon spawning has been reported were sampled for the Upper Kuskokwim River group, though sample sizes for Big River and Middle and Windy Forks were small (N<31, Table 1). Despite the availability of spawning habitat, fewer coho salmon have been observed in the Big

and Middle Forks and the largest concentrations occur in the South Fork Kuskokwim River and tributaries to Highpower Creek (Figure 1; Schwanke et al. 2002).

Uncertainty in baseline allele frequencies either due to sampling effects or temporal variation in allele frequencies was partially ameliorated by the generally large baseline sample sizes and sampling 11 of the 21 locations over multiple years. The effect of temporal variation in baseline allele frequencies is typically not considered as a source of uncertainty in mixed-stock analysis studies (Waples 1990). The majority of coho salmon return to the Kuskokwim River as 4 year olds (e.g., Bue and Martz 2006; Jasper and Molyneaux 2007). Because one year class dominates returns of coho salmon, the accuracy of the 2008 stock composition estimates may be the highest in our sampling years, given that over half the baseline collections were made 4 years prior to this fishery sampling year (Waples 1990). However, though we did find significant variation in temporal samples, temporal variation was by far smaller component of genetic variation in Kuskokwim River coho salmon than spatial variation.

Fishery estimates

Sampling mixtures of coho salmon in the BTF program offered several advantages for estimating the relative proportion and run timing of coho salmon originating from the upper Kuskokwim River in lower river mixtures. Abundance indices generated from the BTF are an important component in management decisions; it is operated daily from June through the end of August, allowing a more complete picture of run timing than would be achieved from commercial catch sampling; and it occurs when and where the majority of commercial- and subsistence catches are made giving an indication of when this stock may be harvested.

Genetic analysis of coho salmon sampled from the BTF resulted in three findings pertinent to fishery management: 1) coho salmon spawning in the upper Kuskokwim River made small contributions to the fishery in the three sampling years, 2) there was a low incidence of Kuskokwim Bay coho salmon in the BTF catches, and 3) Upper Kuskokwim River coho salmon were present in the lower river in throughout the time when commercial fishing occurs, and did not show evidence of earlier run timing during the three years of this study.

Stock proportions (Objective 1)

Coho salmon from the Upper Kuskokwim River were a very small component of the BTF in 2008-2010, with annual estimates ranging from 5-6%. Though small, the contributions were significant; zero was not part of the 95% confidence interval for any of the estimates. Chum salmon are the only other Pacific salmon that spawn in substantial numbers in upper Kuskokwim River tributaries. Similar to coho salmon, upper Kuskokwim River chum salmon spawn later and are genetically distinct from lower river fish (Gilk et al. 2009).   Gilk et al. (2009) sampled chum salmon from a fish wheel at Kalskag from June 14 to September 8 to determine the run timing of chum salmon spawning in the Upper Kuskokwim River. Similar to our coho salmon study, contribution estimates for Upper Kuskokwim River chum salmon for four temporal strata were small, and ranged from 0-5%. These studies indicate that for both species, Upper Kuskokwim River fish are a small proportion of the overall return.

Estensen et al. (2009) reviewed run abundance of coho salmon from 1981-2009. They found that prior to 1996, total run exceeded 1 million fish, while after 1996 abundance has declined by 32%. Estensen et al. (2009) suggested this may be due to anomalous ocean conditions in 1997 and 1998 from which Kuskokwim River coho salmon have not recovered. It would be

interesting to examine scales collected from commercial catch samples before and after this time period to determine if both segments of the populations experienced identical declines in abundance. If not, it could imply that the Upper River stock either has a different marine migration pattern than the Lower River stock, or that other factors besides marine conditions are influencing coho salmon stock abundance in the Kuskokwim River.

Coho salmon harvested within the Kuskokwim River are assumed by managers to be of Kuskokwim River origin, however, results of this study show a small percentage of Kuskokwim Bay coho salmon (3-5%) occasionally enter the Kuskokwim River at least as far as Bethel. It is unclear whether these are strays that would have ultimately spawned in a Kuskokwim River tributary, or exploring fish that would have ultimately returned to spawn in some Kuskokwim Bay tributary. Regardless, these Kuskokwim Bay origin fish could contribute some fraction to the Kuskokwim River harvest. The potential occurrence of interception sometimes becomes an important management issue during times of low salmon run abundance, and this investigation shows that during the years of this study, the occurrence of Kuskokwim Bay fish in the Kuskokwim River upstream of Bethel is likely small.

Timing of Upper Kuskokwim coho salmon (Objective 2)
In contrast to tagging study results (Schaberg et al. 2010), we did not see a pattern of disproportional early timing of river entry for coho spawning in the upper Kuskokwim River. Instead, Upper Kuskokwim coho salmon were consistently present during the entire duration of the BTF, contributing to each quartile of the BTF in each sampling year.

Directed commercial fisheries for coho salmon generally occur from late July until as late as the first week in September (Linderman and Bergstrom 2009). The cumulative catch curves in Figure 5 suggest that in the three years of this study, Upper Kuskokwim River fish were present in the lower river during the entire duration of the commercial fishery, and, at least in 2009 and 2010, likely contributed to commercial catches at levels proportionate to their abundance.

Spawning is timed so that emergence occurs when seasonal environmental conditions are optimum for growth and survival of young (Quinn 2005; Sandercock 1991). Therefore, the timing of freshwater entry is predicted to occur so that fish reach spawning areas within a temporal window that allows for appropriate timing of hatch and fry emergence. In general, earlier migrating fish spawn farther upstream than later migrating fish which enter in a more advanced state of sexual maturity (Weitkamp et al. 1995). The lack of variation in timing of river entry for Kuskokwim River coho salmon stocks suggests that no early adjustment is needed for them to reach their spawning destination, despite longer migration distances. This could indicate that spawners in the upper Kuskokwim River have a later spawning timing, or that the rate of migration is faster for upper river populations, as reported by Schaberg et al. (2010).

Later spawning timing may be due to a different temperature regime in upriver spawning areas. For example, upper Kuskowkim River tributaries are generally turbid from glacial flour, as these streams are fed largely by meltwater from the Alaska Range (Stokes 1985). In aerial surveys, spawning coho salmon were observed in clearwater tributaries and clear side sloughs, whose waters likely originate from groundwater springs (Schwanke and Molyneaux 2002). In the Yukon River, fall-run chum salmon spawn in groundwater upwelling areas, which typically have warmer temperatures than spawning habitats in areas of surface flow, leading to faster embryonic

development (Buklis and Barton 1984). Additionally, smaller egg size is an adaptation in fish with longer spawning migrations, in which case later spawning timing may occur because of more rapid development of smaller eggs (e.g., Gilk et al. 2009).

The largest estimates for Upper Kuskokwim River in the BTF was made in the last quartile of the fishery in every year of the study, but was only statistically larger in 2008. This suggests that though our sample sizes were large enough to be able to detect the presence of Upper Kuskokwim River fish, sample sizes were not large enough to detect small changes in stock composition estimates with certainty. However, the consistent pattern of larger contributions of the Upper Kuskokwim during the last quartile of the fishery in the three years of the study suggests a slightly later return timing for some portions of the Upper Kuskokwim River stock group, either because of later spawning timing as discussed above, or because some portion of the Upper Kuskokwim River group remains at sea for a longer period of time than fish originating from the lower Kuskokwim River.

The majority of growth in salmon is accomplished during the marine portion of their life history. That portion of the Upper Kuskokwim River group with the longest freshwater migrations may be remaining at sea for a few extra weeks to meet the energetic demands of a longer migration, or smaller fish may stay at sea a bit longer to increase their size and reproductive potential. Extra time at sea may be especially critical for coho salmon that only feed at sea for a single year before returning to spawn. Alternatively, coho salmon commonly hold for months in estuarine areas or in freshwater for months before moving to spawning areas (e.g., Halupka et al. 2000). Upper Kuskokwim River fish may be milling in the lower river, and then begin to move

upstream in increasing numbers later in the BTF, giving the appearance of a slightly later run timing.

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DELIVERABLES:

The project deliverables from this study are 1) a final report and six semiannual reports available

from the AYK SSI program or through the Conservation Genetics Laboratory, U.S. Fish and

Wildlife Service; 2) oral presentations made at the Alaska Chapter American Fisheries Society

meeting in November 2008 and the Spring Kuskokwim Interagency Meetings in March of 2009,

2010, and 2011; 3) an archive of tissue samples and an Excel spreadsheet of genotypes for coho

salmon collected through this project (see Table 1; tissue samples and genetic data are available

upon request from the Conservation Genetics Laboratory); 5) a manuscript is in preparation for

submission to the Journal of Fish and Wildlife Management in 2013); and a poster is in

preparation summarizing project results to be given to the Orutsararmiut Native Council (ONC)

and Alaska Department of Fish and Game (ADFG) offices in Bethel.

PROJECT DATA:

Tissue samples for coho salmon collected in this study are stored in 2ml cryovials and preserved

in ethanol. Associated collection information for samples is archived in a Microsoft Access

database and genotypic data are maintained in an Excel spreadsheet (Microsoft Office 2010).

Tissue subsamples and data are available upon request to John Wenburg, Conservation Genetics

Laboratory, U.S. Fish and Wildlife Service, 1011 E. Tudor Road, Anchorage, AK, USA 99503.

Phone (907) 786-3858, email: john_wenburg@fws.gov.

ACKNOWLEDGEMENTS:

PRESS RELEASE:

Coho salmon spawning in the upper portion of the Kuskokwim River upstream of the Takotna River represent an important component of the genetic and life history variation for coho salmon in the watershed. Mark-recapture studies conducted in the Kuskokwim River suggest that coho salmon spawning in tributaries more distal from river mouth have progressively earlier run timing. Early run timing can pose a management concern because information to assess run

abundance is the least reliable early in the fishing season resulting in a greater risk

overharvesting stocks with early run timing. In addition, if overharvest does occur in stocks

spawning in the upper Kuskokwim River, there are no stock assessment projects to evaluate the

impacts on escapement upstream of the Takotna River. In this study, coho salmon were sampled

from a test fishery conducted annually near Bethel in 2008-2010. Mixed-stock analysis using

genetic characters was used to estimate the contributions of three stock groups to the test fishery

samples (Upper Kuskokwim River, coho salmon spawning in tributaries upstream of the Takotna

River; Lower Kuskokwim River, coho salmon spawning in tributaries from the river mouth to

the Takotna River; and Kuskokwim Bay, coho salmon spawning in the Middle Fork Goodnews,

Arolik, and Kanektok rivers) to determine their relative abundance in the test fishery over a

three-year period and to determine if Upper Kuskokwim River coho salmon have an earlier run

timing compared to stocks in the lower and middle river. Findings from this study pertinent to

fishery management are that 1) coho salmon spawning in the upper Kuskokwim River occurred

in low abundance in test fishery in all sampling years, with an annual average contribution of 5-

6%, 2) they were present in the lower river throughout the time when commercial fishing occurs,

and did not show evidence of earlier run timing than other Kuskokwim River stocks during the

three years of this study, and 3) there was a low incidence of Kuskokwim Bay coho salmon in

the test fishery catches.
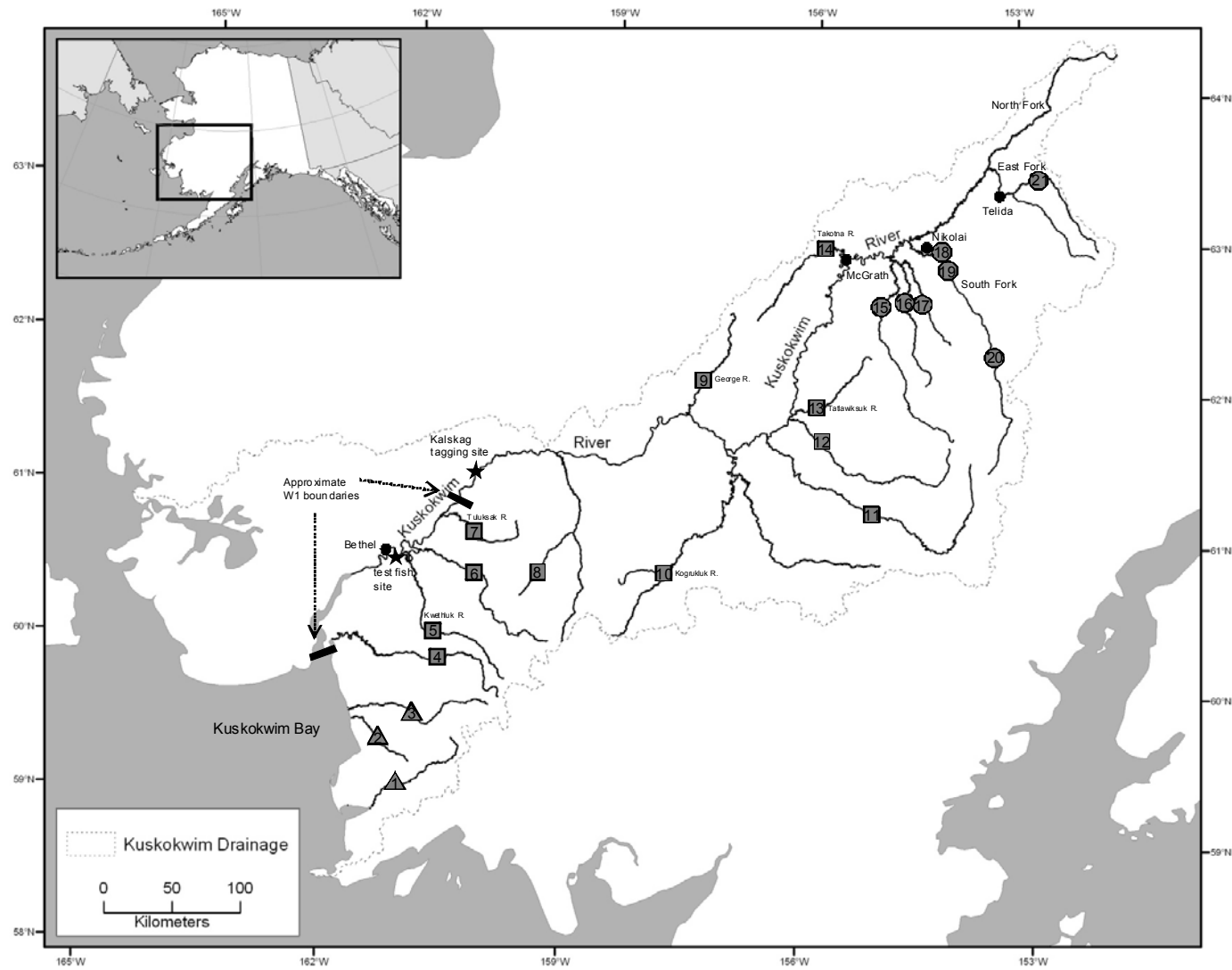
Figure 1. Sampling locations for coho salmon *Oncorhynchus kisutch* collected in the Kuskokwim River area, Alaska, from 1997-2010 to estimate the stock composition of coho salmon sampled from the Bethel test fishery. Symbols represent stock groupings used in the fishery estimates: triangles, Kuskokwim Bay; squares, Lower Kuskokwim River; circles, Upper Kuskokwim River. Numbers correspond to sampling locations described in Table 1.
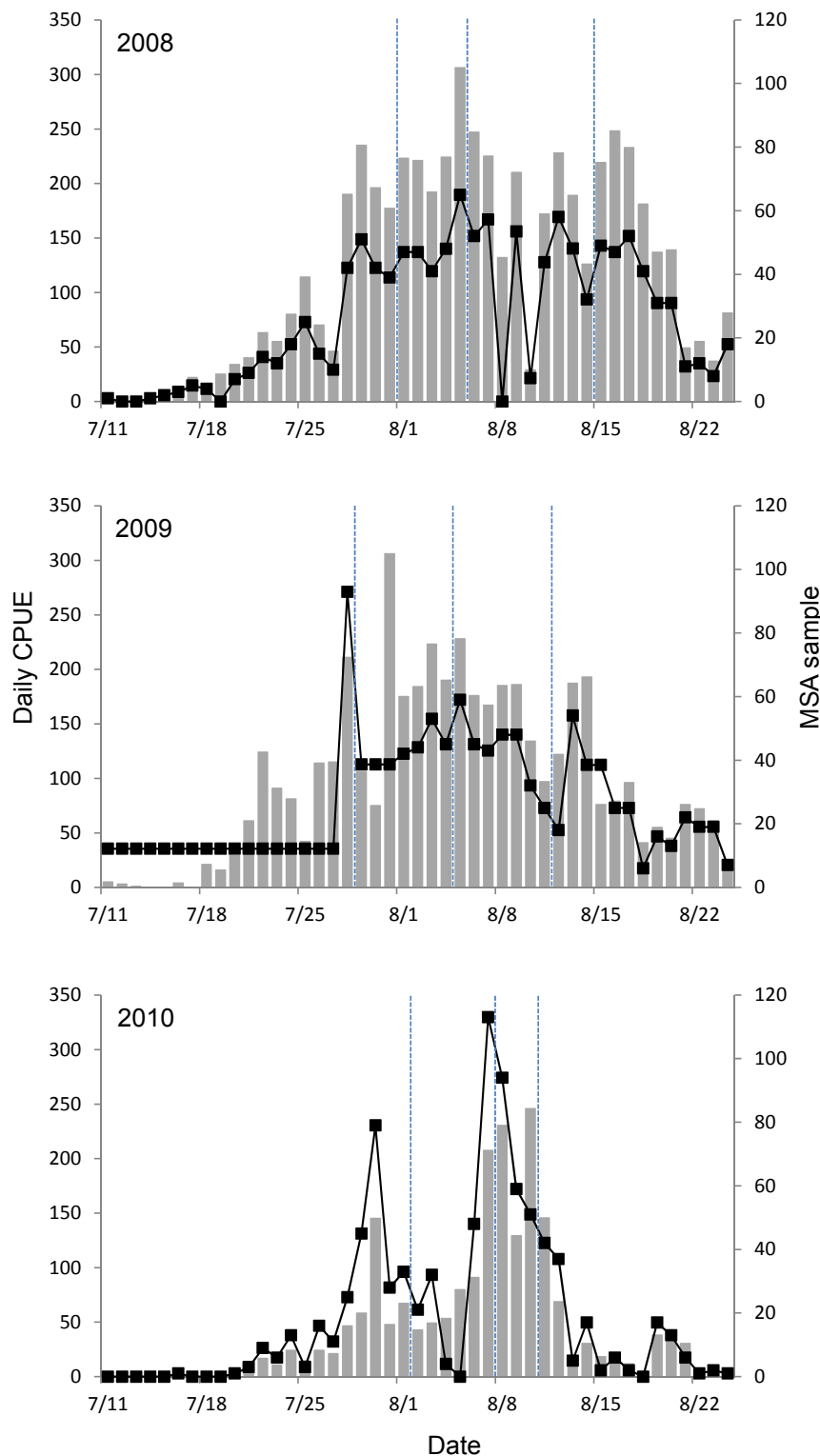
Figure 2. The stock composition for coho salmon *Oncorhynchus kisutch* was estimated for each quartile of the Bethel test fishery in the Kuskokwim River, Alaska, from 2008-2010. For each estimate, N=300 fish were randomly subsampled proportional to the daily CPUE in each quartile. In 2010, all fish collected from the fishery were analyzed because of low catch rates.
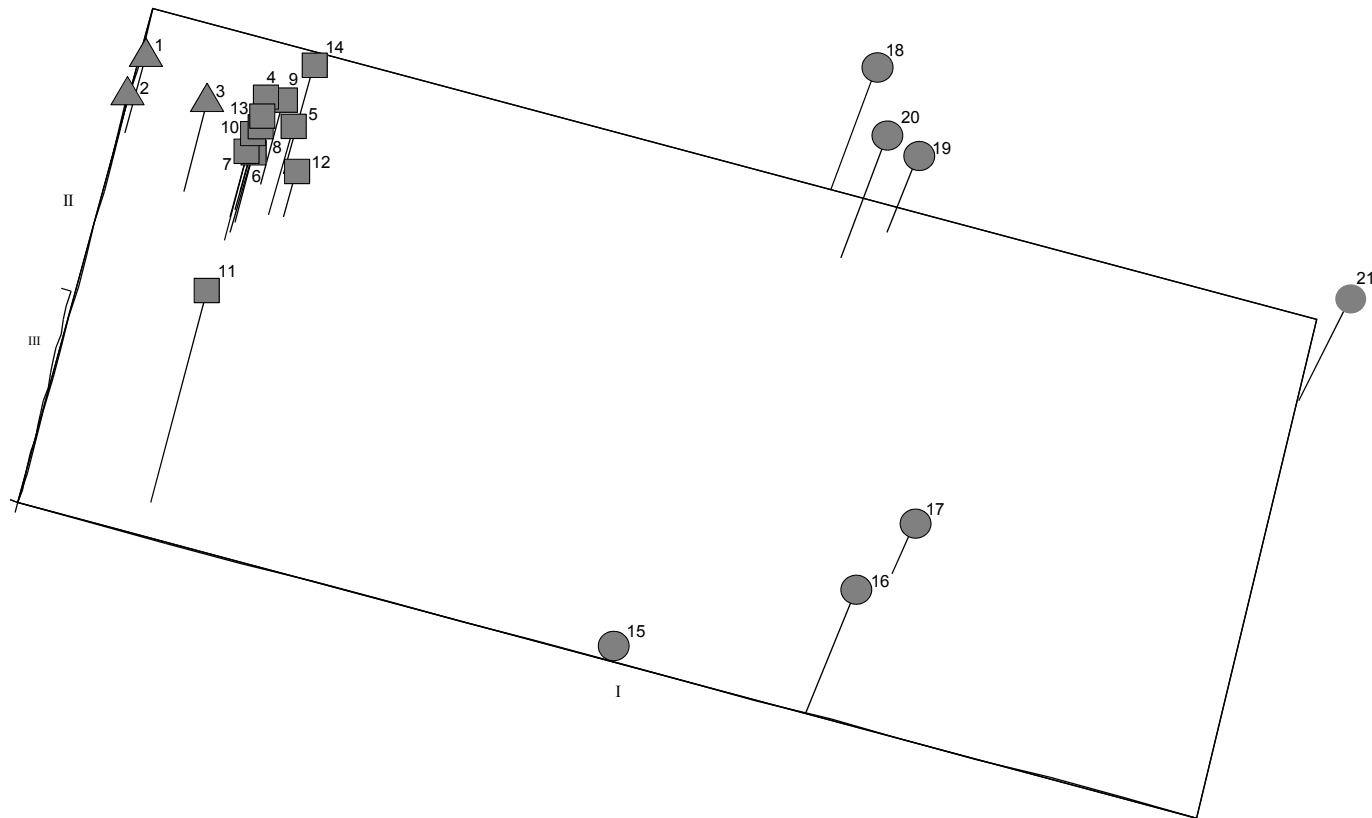
Figure 3. Multidimensional scaling analysis of coho salmon *Oncorhynchus kisutch* collected in the Kuskokwim River area, Alaska from 1997-2010. Numbers correspond to sampling locations in Figure 1 and Table 1. Symbols represent stock groupings used in the fishery estimates: triangles, Kuskokwim Bay; squares, Lower Kuskokwim River; circles, Upper Kuskokwim River.
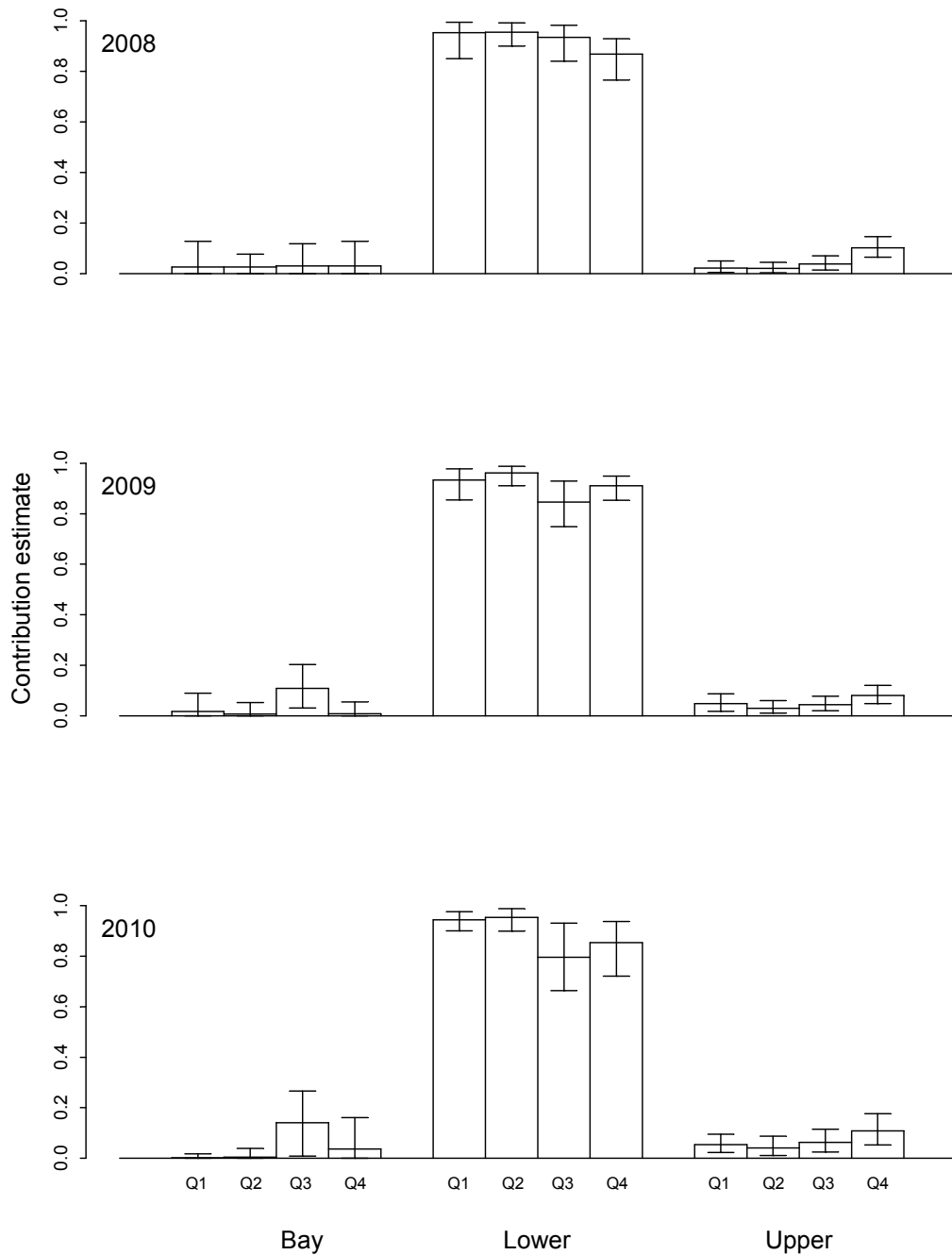
Figure 4. Estimates for three stock groups of coho salmon *Oncorhynchus kisutch* (Upper Kuskokwim River, Lower Kuskokwim River, and Kuskokwim Bay) for each quartile of the 2008, 2009, and 2010 Bethel test fishery in the lower Kuskokwim River, Alaska. In each year, the largest contribution estimate for the Upper Kuskokwim occurred in the last quartile. Error bars represent 95% credibility intervals.
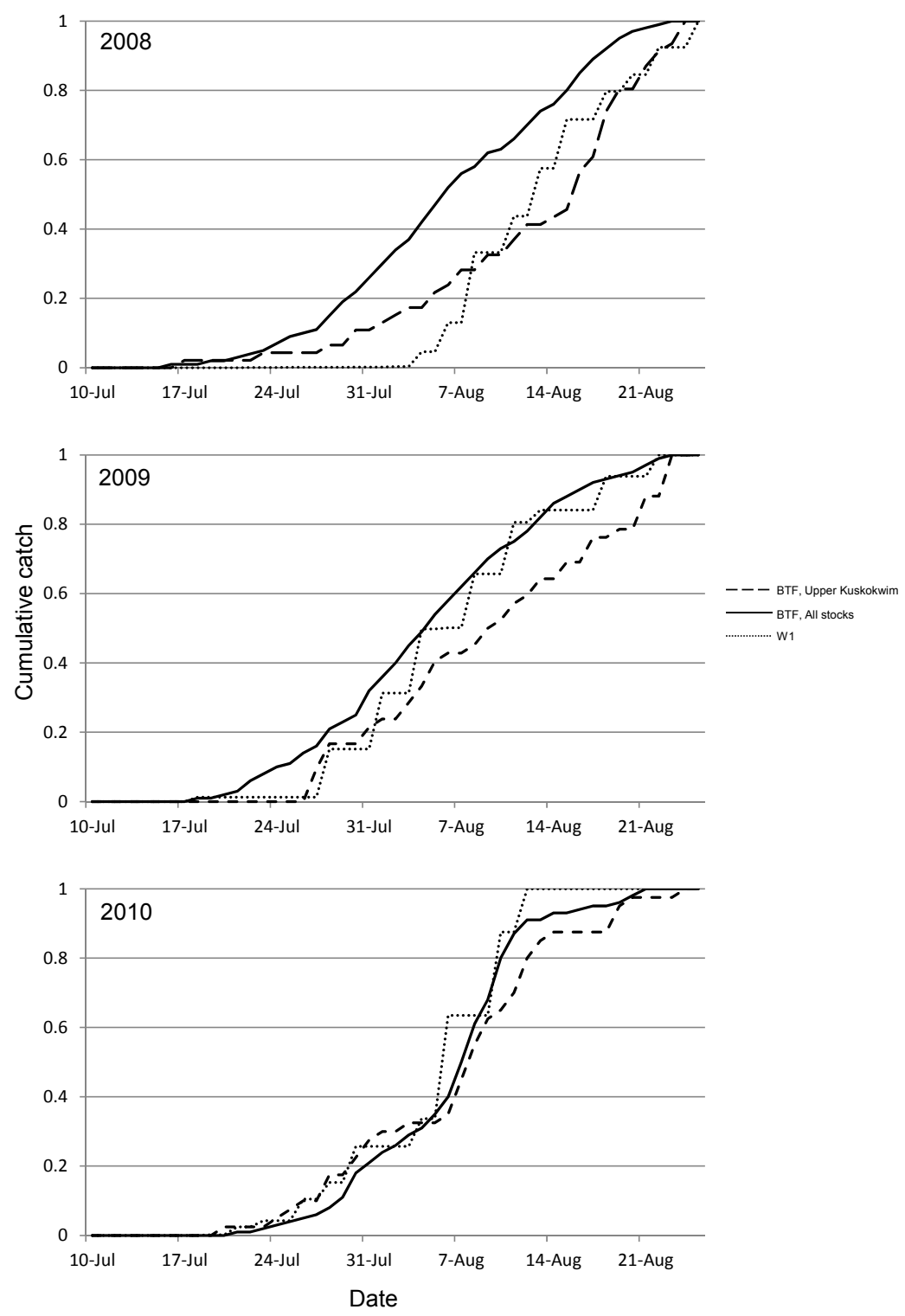
Figure 5. Cumulative catch curves for coho salmon *Oncorhynchus kisutch* in the Bethel test fishery and District W1 commercial catches in the Kuskokwim River, Alaska from 2008-2010. Individual-based analysis was used to derive cumulative catch curves for coho salmon from the Upper Kuskokwim River.

Table 1. Location, date, and sample size of baseline and fishery samples collected to estimate the contribution of three stock groups of coho salmon *Oncorhynchus kistuch* in the Bethel test fishery in the lower Kuskokwim River, Alaska in 2008-2010. Map numbers correspond to Figure 1.

Collection Type	Stock Groups	Location	Map No.	Year collected	N
Baseline	Kuskokwim Bay	Middle Fork Goodnews River (weir)	1	2004	200
				2001	95
		Arolik River	2	1997	83
		Kanektok River (weir)	3	2004	100
				2001	96
	Lower Kuskokwim R.	Eek River (juveniles)	4	2010	168
		Kwethluk River (weir)	5	2004	200
		Kisaralik River	6	2004	55
				1997	82
		Tuluksak River (weir)	7	2004	200
		Salmon River	8	2007	197
		Salmon River (weir)		2004	100
		George River (weir)	9	2004	100
				2001	96
		Kogrukluk River	10	2004	100
				2001	96
		Stony River	11	2008	27
		Stony River (radio tracked)		2008	12
		Swift River (Cheeneetnuk River)	12	2009	110
		Swift River (radio tracked)		2008	61
		Tatlawiksuk River	13	2004	100
				2001	96
		Takotna River	14	2004	100
				2001	96
	Upper Kuskokwim R.	Big River	15	2008	23
		Middle Fork	16	2008	21
		Middle Fork (radio-tracked)		2008	10
		Windy Fork	17	2008	27
		South Fork	18	2008	65
			19	2004	200
		South Fork (Tin Creek)	20	2008	120
		Highpower Creek	21	2005	21
				2004	29
Mixture		2008 Bethel test fishery, 11 July-11 Sept.		2008	2562
		2009 Bethel test fishery, 11 July-24 Aug.		2009	1846
		2010 Bethel test fishery, 16 July-24 Aug.		2010	846

Table 1. Continued.

Collection Type	Stock Groups	Location	Map No.	Year collected	N
Mixture		2008 W1, August 4		2008	408
		2008 W1, August 6		2008	319
		2008 W1, August 11		2008	438
		2008 W1, August 13		2008	469
		2008 W1, August 18		2008	476
		2008 W1, August 20		2008	428
		2008 W1, August 25		2008	447
		2009 W1, July 18		2009	5
		2009 W1, July 28		2009	164
		2009 W1, August 1		2009	543
		2009 W1, August 4		2009	150
		2009 W1, August 8		2009	400
		2009 W1, August 18		2009	254
		2009 W1, August 22		2009	469
		2010 W1A, July 28		2010	407
		2010 W1A, August 4		2010	456
		2010 W1A, August 10		2010	414

Table 2. Mean stock group estimates and 95% credibility intervals for mixtures of coho salmon *Oncorhynchus kisutch* from the Kuskokwim River area, Alaska. Mixtures are of known origin, and were created by deleting genotypes from the baseline data set, and estimating stock contributions with a reduced baseline. Stock groups are defined in Table 1 and Figure 1, and mixtures were estimated using the program Bayes (Pella and Masuda 2001).

Mixture	Stock Group (Known Proportion)	Mean	S.D.	Credibility Interval 2.5%	97.5%
1	Upper Kuskokwim (1)	0.993	0.008	0.972	1.000
	Lower Kuskokwim (0)	0.006	0.007	0.000	0.026
	Kuskokwim Bay (0)	0.001	0.003	0.000	0.011
2	Upper Kuskokwim (0)	0.005	0.007	0.000	0.025
	Lower Kuskokwim (1)	0.960	0.040	0.861	1.000
	Kuskokwim Bay (0)	0.036	0.040	0.000	0.133
3	Upper Kuskokwim (0)	0.002	0.004	0	0.0123
	Lower Kuskokwim (0)	0.058	0.051	0.0003	0.1811
	Kuskokwim Bay (1)	0.940	0.051	0.8166	0.9991
4	Upper Kuskokwim (0.02)	0.022	0.012	0.004	0.050
	Lower Kuskokwim (0.91)	0.888	0.035	0.811	0.947
	Kuskokwim Bay (0.07)	0.090	0.033	0.036	0.165

Table 3. Stock composition estimates for coho salmon *Oncorhynchus kisutch* sampled from each quartile of the Bethel test fishery in the Kuskokwim River, Alaska in 2008-2010. Estimates were computed in Bayes (Pella and Masuda 2001).

Year	Quartile	Stock Group	Mean	S.D.	Credibility Interval 2.5%	97.5%
2008	Quartile 1	Upper Kuskokwim	0.021	0.012	0.004	0.049
		Lower Kuskokwim	0.953	0.039	0.850	0.994
		Kuskokwim Bay	0.026	0.037	0.000	0.127
	Quartile 2	Upper Kuskokwim	0.020	0.011	0.003	0.044
		Lower Kuskokwim	0.955	0.024	0.899	0.991
		Kuskokwim Bay	0.025	0.022	0.000	0.077
	Quartile 3	Upper Kuskokwim	0.037	0.014	0.014	0.070
		Lower Kuskokwim	0.934	0.039	0.841	0.982
		Kuskokwim Bay	0.029	0.036	0.000	0.117
	Quartile 4	Upper Kuskokwim	0.102	0.021	0.064	0.146
		Lower Kuskokwim	0.868	0.042	0.766	0.929
		Kuskokwim Bay	0.030	0.037	0.000	0.127
2009	Quartile 1	Upper Kuskokwim	0.048	0.018	0.018	0.087
		Lower Kuskokwim	0.934	0.032	0.855	0.978
		Kuskokwim Bay	0.018	0.026	0.000	0.090
	Quartile 2	Upper Kuskokwim	0.030	0.013	0.011	0.061
		Lower Kuskokwim	0.962	0.020	0.911	0.988
		Kuskokwim Bay	0.008	0.015	0.000	0.053
	Quartile 3	Upper Kuskokwim	0.045	0.015	0.020	0.077
		Lower Kuskokwim	0.846	0.046	0.748	0.929
		Kuskokwim Bay	0.110	0.044	0.031	0.203

Table 3. Continued.

Year	Quartile	Stock Group	Mean	S.D.	2.5%	97.5%
					Credibility Interval	
2009	Quartile 4	Upper Kuskokwim	0.081	0.019	0.048	0.121
		Lower Kuskokwim	0.911	0.024	0.854	0.949
		Kuskokwim Bay	0.008	0.016	0.000	0.055
2010						
	Quartile 1	Upper Kuskokwim	0.054	0.019	0.023	0.095
		Lower Kuskokwim	0.944	0.019	0.901	0.976
		Kuskokwim Bay	0.002	0.006	0.000	0.018
	Quartile 2	Upper Kuskokwim	0.042	0.020	0.011	0.088
		Lower Kuskokwim	0.954	0.023	0.900	0.988
		Kuskokwim Bay	0.005	0.011	0.000	0.039
	Quartile 3	Upper Kuskokwim	0.063	0.023	0.025	0.115
		Lower Kuskokwim	0.796	0.067	0.663	0.931
		Kuskokwim Bay	0.141	0.062	0.008	0.266
	Quartile 4	Upper Kuskokwim	0.109	0.032	0.053	0.177
		Lower Kuskokwim	0.854	0.056	0.721	0.937
		Kuskokwim Bay	0.038	0.047	0.000	0.162

APPENDICES:

Appendix 1. Sample size (N), allele frequencies, and p-values (P-HW) of genotypic frequencies to Hardy-Weinberg expectation for coho salmon *Oncorhynchus kisutch* from the Kuskokwim River area, Alaska. Population numbers are described in Table 1.

Locus		Population																				
		1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21
Ocl8																							
N		292	79	192	167	194	131	196	291	191	194	38	170	193	191	23	31	27	199	65	120	50
	94	0	0	0	0	0	0.004	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
	96	0	0	0	0	0	0	0.003	0.003	0	0	0	0	0	0	0	0	0	0	0	0	0
	98	0.019	0.082	0.07	0.042	0.085	0.073	0.082	0.082	0.052	0.082	0.158	0.026	0.047	0.076	0.109	0.113	0.056	0.048	0.108	0.087	0.01
	100	0.002	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
	102	0.063	0.019	0.029	0.018	0.021	0.015	0.013	0.014	0.026	0.01	0.026	0.012	0	0.01	0	0	0	0.003	0.023	0.004	0
	104	0.021	0.019	0.018	0.009	0.008	0.011	0.01	0.005	0.005	0.01	0	0.065	0.003	0.071	0	0.016	0	0	0	0	0.01
	106	0.601	0.614	0.615	0.689	0.647	0.618	0.625	0.643	0.654	0.608	0.526	0.738	0.725	0.647	0.565	0.548	0.537	0.781	0.746	0.758	0.77
	108	0.122	0.095	0.125	0.117	0.111	0.16	0.128	0.129	0.136	0.157	0.013	0.026	0.132	0.105	0.043	0	0.056	0.023	0	0.004	0
	110	0.021	0.013	0.018	0.012	0.013	0.008	0.013	0.005	0.003	0.005	0.092	0.015	0.005	0.013	0.087	0.129	0.074	0.083	0.077	0.071	0.05
	112	0	0	0	0	0.005	0	0.005	0	0.003	0	0	0	0	0	0	0	0	0	0	0	0
	114	0.003	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
	116	0	0	0	0	0	0	0.003	0	0	0	0	0	0	0	0	0	0	0	0	0	0.01
	120	0.046	0.051	0.055	0.036	0.049	0.073	0.079	0.076	0.063	0.085	0.171	0.1	0.044	0.045	0.065	0.177	0.204	0.028	0.038	0.05	0.15
	124	0	0.006	0.003	0.003	0.005	0	0.005	0.007	0.026	0.01	0	0.003	0	0	0.109	0	0	0	0	0	0
	126	0.002	0	0	0	0	0	0	0	0	0	0	0	0	0.003	0	0	0	0	0	0	0
	136	0.005	0	0	0.006	0	0.004	0	0.002	0.003	0.003	0.013	0.006	0.003	0.003	0	0	0	0	0	0.008	0
	138	0.005	0.006	0	0	0.008	0.008	0	0.002	0	0	0	0	0	0	0	0	0	0.008	0	0	0
	140	0.089	0.095	0.06	0.063	0.044	0.027	0.036	0.033	0.029	0.028	0	0.009	0.041	0.026	0	0	0	0.028	0.008	0.017	0
	142	0	0	0.003	0	0.003	0	0	0	0	0	0	0	0	0.003	0	0	0	0	0	0	0
	144	0.002	0	0.005	0.006	0.003	0	0	0	0	0	0	0	0	0	0.022	0.016	0.074	0	0	0	0
P-HW		0.692	0.489	0.842	0.197	0.671	0.628	0.257	0.419	0.564	0.638	0.806	0.237	0.461	0.032	0.515	0.100	0.775	0.634	0.530	0.006	0.501
Oke2																							
N		288	78	194	168	200	135	198	296	195	196	38	171	196	194	23	31	27	200	65	120	49
	167	0.885	0.885	0.879	0.923	0.902	0.904	0.874	0.892	0.885	0.898	0.934	0.912	0.885	0.899	1	1	0.963	0.885	0.962	0.975	1

Appendix 1. Continued.

Locus										Population											
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21
Oke2																					
169	0.115	0.115	0.119	0.077	0.098	0.096	0.126	0.108	0.115	0.102	0.066	0.088	0.115	0.101	0	0	0.037	0.115	0.038	0.025	0
171	0	0	0.003	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
P-HW	0.396	1.000	0.546	0.602	0.226	0.610	1.000	0.551	1.000	0.699	1.000	0.620	1.000	1.000			1.000	0.082	1.000	1.000	
Oke3																					
N	294	80	187	167	197	130	192	290	190	193	38	171	192	190	23	31	27	194	65	120	45
251	0.019	0.031	0.035	0.039	0.063	0.054	0.068	0.043	0.047	0.047	0.092	0.015	0.049	0.029	0	0.016	0.019	0.005	0	0	0
253	0	0	0	0	0.003	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
255	0.019	0.006	0.011	0.006	0.015	0.027	0.005	0.021	0.016	0.016	0	0	0.003	0.005	0	0	0	0.013	0.008	0.004	0.044
261	0	0	0	0	0.003	0	0	0	0	0	0	0	0	0	0	0	0	0.003	0	0	0
263	0.272	0.225	0.246	0.231	0.135	0.162	0.211	0.186	0.221	0.244	0.382	0.263	0.286	0.237	0.391	0.194	0.148	0.193	0.138	0.175	0.144
267	0.002	0	0	0	0	0	0.005	0.002	0	0	0	0	0	0	0	0	0	0	0	0	0
271	0.003	0	0.003	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
273	0.005	0	0	0	0	0	0.003	0	0.021	0	0	0	0	0	0	0	0	0	0	0	0
275	0.522	0.631	0.567	0.572	0.581	0.562	0.547	0.636	0.513	0.557	0.5	0.48	0.529	0.647	0.522	0.565	0.574	0.606	0.631	0.6	0.7
277	0.158	0.106	0.139	0.153	0.201	0.196	0.161	0.112	0.179	0.137	0.026	0.243	0.133	0.082	0.087	0.226	0.259	0.18	0.223	0.221	0.111
285	0	0	0	0	0	0	0	0	0.003	0	0	0	0	0	0	0	0	0	0	0	0
P-HW	0.206	0.610	0.286	0.211	0.876	0.121	0.412	0.489	0.077	0.666	0.776	0.636	0.658	0.094	0.069	0.895	0.268	0.615	0.533	0.733	0.054
Oke4																					
N	295	78	194	167	199	132	196	293	194	196	38	171	195	194	23	31	27	182	65	119	49
232	0	0	0	0	0.01	0	0.015	0.003	0.003	0.008	0	0.006	0.003	0.008	0	0	0	0	0	0	0
236	0.086	0.045	0.046	0.03	0.01	0.023	0.023	0.024	0.067	0.033	0	0.023	0.018	0.049	0	0	0	0.005	0	0	0
240	0.91	0.942	0.954	0.964	0.98	0.977	0.962	0.968	0.92	0.959	1	0.971	0.979	0.943	1	1	1	0.995	1	1	1
242	0.003	0	0	0.003	0	0	0	0.005	0.01	0	0	0	0	0	0	0	0	0	0	0	0
244	0	0.013	0	0.003	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
P-HW	0.758	1.000	1.000	1.000	1.000	1.000	1.000	1.000	0.728	1.000		0.051	1.000	1.000				1.000			

Appendix 1. Continued.

										Population										
Locus	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21
Oki1																					
N	294	79	194	167	199	136	197	295	192	196	38	171	195	194	23	31	27	200	65	120	49
90	0.002	0	0.003	0.009	0	0	0	0	0.008	0.003	0	0.02	0	0	0	0.016	0	0.003	0	0	0
94	0.027	0.019	0.013	0.015	0.02	0.011	0.02	0.025	0.031	0.026	0	0.009	0.015	0.028	0	0	0	0.015	0.015	0.013	0.02
98	0.447	0.437	0.479	0.434	0.43	0.46	0.477	0.461	0.492	0.515	0.224	0.57	0.454	0.472	0.283	0.323	0.315	0.242	0.215	0.308	0.296
102	0.221	0.158	0.165	0.135	0.163	0.077	0.107	0.125	0.13	0.112	0.276	0.146	0.146	0.142	0.239	0.242	0.204	0.175	0.246	0.158	0.347
106	0.085	0.139	0.106	0.165	0.138	0.202	0.162	0.142	0.133	0.089	0.145	0.161	0.128	0.165	0.348	0.21	0.352	0.472	0.415	0.321	0.327
110	0.175	0.196	0.219	0.231	0.236	0.232	0.221	0.239	0.195	0.237	0.355	0.082	0.254	0.188	0.065	0.048	0.074	0.075	0.108	0.192	0.01
114	0.027	0.032	0.015	0.009	0.013	0.011	0.013	0.007	0.01	0.018	0	0.009	0.003	0.005	0.065	0.161	0.056	0.018	0	0.008	0
118	0.003	0.006	0	0.003	0	0.007	0	0	0	0	0	0.003	0	0	0	0	0	0	0	0	0
122	0.01	0.013	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
130	0.002	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
P-HW	0.426	0.700	0.676	0.468	0.010	0.188	0.443	0.645	0.721	0.931	0.042	0.327	0.189	0.297	0.682	0.146	0.135	0.323	0.718	0.191	0.780
Oki11																					
N	295	81	194	168	200	135	199	295	195	195	38	171	183	188	23	31	27	200	65	120	50
82	0.773	0.772	0.794	0.854	0.813	0.819	0.791	0.834	0.846	0.859	0.671	0.822	0.85	0.864	0.87	0.871	0.833	0.98	0.954	0.95	0.99
84	0	0	0	0	0	0	0	0	0.003	0	0	0	0.003	0	0	0	0	0	0	0	0
86	0.225	0.228	0.206	0.146	0.172	0.17	0.198	0.151	0.136	0.136	0.329	0.178	0.145	0.136	0.13	0.129	0.167	0.02	0.046	0.05	0.01
88	0.002	0	0	0	0.015	0.011	0.01	0.015	0.015	0.005	0	0	0.003	0	0	0	0	0	0	0	0
P-HW	0.030	0.545	0.270	0.534	0.451	1.000	0.929	0.331	0.039	0.244	0.483	0.068	0.100	0.346	1.000	0.403	0.549	1.000	1.000	1.000	
Oki3																					
N	294	81	195	168	196	134	197	293	193	193	38	171	187	194	23	31	27	187	65	120	50
60	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0.003	0	0	0
63	0	0	0	0	0	0	0.01	0	0	0	0	0	0	0	0	0	0	0	0	0	0
66	0.172	0.173	0.203	0.28	0.293	0.28	0.256	0.249	0.295	0.303	0.368	0.418	0.297	0.376	0.283	0.645	0.556	0.655	0.608	0.629	0.78
69	0.828	0.827	0.797	0.72	0.704	0.716	0.731	0.751	0.705	0.697	0.592	0.582	0.703	0.624	0.717	0.355	0.444	0.342	0.392	0.371	0.22

Appendix 1. Continued.

	Population																				
Locus	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21
Oki3																					
72	0	0	0	0	0.003	0.004	0.003	0	0	0	0.039	0	0	0	0	0	0	0	0	0	0
P-HW	0.098	0.238	0.825	0.565	0.508	0.658	0.739	0.041	0.015	0.495	0.880	0.117	0.293	0.221	0.626	0.242	0.444	0.617	1.000	0.174	0.214
Omy1011																					
N	288	79	195	165	191	134	184	293	196	194	38	171	192	194	23	31	27	183	65	120	50
174	0	0	0	0	0	0	0	0	0.003	0	0	0	0	0	0	0	0	0	0	0	0
178	0.247	0.247	0.269	0.267	0.275	0.284	0.242	0.251	0.332	0.253	0.171	0.289	0.214	0.296	0.087	0.468	0.185	0.221	0.323	0.321	0.31
182	0.226	0.19	0.208	0.158	0.157	0.149	0.147	0.133	0.166	0.162	0.132	0.161	0.188	0.173	0.522	0.387	0.593	0.607	0.362	0.479	0.63
186	0.17	0.234	0.182	0.218	0.225	0.209	0.253	0.246	0.189	0.284	0.197	0.199	0.297	0.26	0.217	0.065	0.093	0.066	0.138	0.071	0.06
190	0	0	0	0	0	0.004	0.003	0.002	0	0.003	0	0	0	0	0	0	0.056	0	0	0.004	0
194	0.007	0	0.005	0	0	0	0	0	0	0.003	0	0.003	0.003	0	0	0	0	0	0.008	0	0
198	0.057	0.032	0.026	0.012	0.024	0.011	0.011	0.019	0.036	0.008	0	0	0.01	0.049	0.022	0	0	0	0	0	0
202	0.005	0.006	0.01	0.003	0.003	0	0.003	0.002	0	0.008	0	0.006	0	0	0	0	0	0	0	0	0
206	0.068	0.07	0.074	0.1	0.097	0.078	0.079	0.073	0.066	0.07	0.158	0.056	0.078	0.106	0.043	0	0	0.025	0.038	0.033	0
210	0.155	0.158	0.162	0.152	0.162	0.19	0.168	0.181	0.151	0.139	0.276	0.205	0.125	0.08	0.043	0.081	0.019	0.057	0.115	0.083	0
214	0.012	0.019	0.023	0.042	0.031	0.026	0.049	0.026	0.026	0.021	0	0.023	0.021	0.021	0	0	0.019	0.014	0.015	0.004	0
218	0.049	0.044	0.041	0.045	0.024	0.049	0.038	0.065	0.028	0.046	0.066	0.056	0.065	0.015	0.065	0	0.037	0.011	0	0.004	0
222	0.005	0	0	0.003	0.003	0	0.005	0.003	0.003	0.005	0	0	0	0	0	0	0	0	0	0	0
230	0	0	0	0	0	0	0.003	0	0	0	0	0	0	0	0	0	0	0	0	0	0
234	0	0	0	0	0	0	0	0	0.003	0	0	0.003	0	0	0	0	0	0	0	0	0
P-HW	0.852	0.655	0.035	0.745	0.811	0.502	0.511	0.092	0.019	0.119	0.045	0.252	0.584	0.015	0.478	0.561	0.607	0.092	0.251	0.162	0.108
Oneu3																					
N	295	71	192	163	197	135	198	290	193	190	38	171	189	193	23	31	27	199	65	119	48
164	0	0.014	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
168	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0.032	0.019	0	0	0	0
172	0.308	0.394	0.424	0.423	0.409	0.463	0.455	0.453	0.43	0.416	0.526	0.462	0.434	0.427	0.522	0.371	0.481	0.583	0.508	0.437	0.563

Appendix 1. Continued.

Locus	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21
																		Population			
Oneu3																					
174	0.219	0.169	0.18	0.221	0.193	0.185	0.179	0.207	0.223	0.208	0.105	0.304	0.161	0.14	0.261	0.306	0.315	0.284	0.254	0.328	0.24
176	0.002	0	0	0	0	0	0.003	0	0	0	0	0	0	0	0	0	0	0	0	0	0
178	0.207	0.254	0.211	0.23	0.254	0.207	0.197	0.2	0.22	0.229	0.197	0.108	0.228	0.303	0.174	0.274	0.185	0.106	0.169	0.181	0.188
180	0	0	0	0.003	0	0.004	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
182	0.264	0.169	0.185	0.12	0.145	0.141	0.167	0.14	0.127	0.147	0.171	0.126	0.177	0.127	0.043	0.016	0	0.028	0.069	0.055	0.01
192	0	0	0	0	0	0	0	0	0	0	0	0	0	0.003	0	0	0	0	0	0	0
196	0	0	0	0.003	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
P-HW	0.729	0.036	0.446	0.236	0.312	0.309	0.832	0.446	0.643	0.791	0.079	0.628	0.632	0.064	0.043	0.323	0.061	0.009	0.816	0.355	0.870
Ots101																					
N	275	81	193	168	196	132	186	294	192	195	38	171	190	192	23	31	27	179	65	120	50
98	0	0	0	0	0	0	0	0	0	0	0	0	0.003	0	0	0	0	0	0	0	0
106	0.015	0.025	0.016	0.006	0.003	0	0	0	0.008	0	0	0	0.003	0.003	0	0	0	0.003	0.015	0	0
110	0.027	0.019	0.028	0.015	0.008	0.011	0.008	0.024	0.01	0.031	0.053	0.023	0.026	0.01	0	0	0	0	0	0	0
114	0.002	0.006	0.023	0	0	0	0	0.005	0	0.003	0	0.009	0.003	0.005	0	0	0	0	0.008	0	0
126	0.004	0	0.003	0.015	0.003	0.019	0.005	0.012	0	0	0	0	0	0	0	0.016	0	0	0	0	0
130	0.013	0.012	0.008	0.003	0	0	0	0.005	0.003	0	0	0	0	0	0	0	0	0	0	0	0
134	0.033	0.037	0.021	0.006	0.015	0.008	0	0	0.005	0.005	0	0	0	0.005	0	0	0	0	0	0	0
138	0.022	0.006	0.01	0	0.005	0.008	0.013	0.015	0.01	0.008	0	0.003	0.011	0.003	0	0	0	0	0	0	0
142	0.047	0.019	0.052	0.009	0.031	0.015	0.032	0.022	0.047	0.038	0.079	0.047	0.061	0.042	0	0	0.037	0.014	0	0	0
146	0.109	0.062	0.078	0.074	0.097	0.064	0.099	0.077	0.094	0.108	0.013	0.07	0.084	0.091	0.174	0.065	0.13	0.025	0.015	0.017	0
150	0.042	0.037	0.052	0.086	0.061	0.072	0.056	0.037	0.091	0.059	0.026	0.067	0.063	0.109	0.022	0.016	0	0.05	0.146	0.113	0.01
154	0.091	0.068	0.073	0.06	0.066	0.061	0.099	0.083	0.052	0.056	0.066	0.132	0.042	0.013	0.022	0	0	0.008	0.015	0.004	0
158	0.069	0.056	0.047	0.051	0.051	0.057	0.027	0.051	0.026	0.031	0.105	0.061	0.032	0.016	0.13	0.081	0	0.022	0.054	0.021	0
162	0.035	0.037	0.018	0.018	0.013	0.015	0	0.005	0.003	0.013	0	0.023	0.024	0.008	0	0.016	0.019	0.003	0.008	0.013	0.05
166	0.024	0.043	0.031	0.009	0.013	0.023	0.011	0.009	0.01	0.01	0.026	0.009	0.003	0.005	0.087	0.129	0.037	0.078	0.023	0.008	0.19
170	0.016	0.043	0.003	0	0.01	0	0.008	0.01	0	0.005	0	0.018	0.016	0.008	0.022	0	0.019	0.045	0.038	0.021	0.03

Appendix 1. Continued.

Locus	Population																				
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21
Ots101																					
174	0.025	0.037	0.041	0.083	0.074	0.064	0.046	0.037	0.086	0.067	0.026	0.056	0.118	0.185	0	0.032	0.074	0.087	0.115	0.15	0.06
178	0.056	0.049	0.054	0.077	0.046	0.072	0.083	0.061	0.07	0.067	0.013	0.041	0.071	0.065	0.152	0	0.019	0.059	0.1	0.079	0.07
182	0.049	0.099	0.088	0.051	0.079	0.053	0.11	0.058	0.081	0.062	0.184	0.029	0.053	0.107	0.043	0.016	0.093	0.073	0.077	0.142	0.08
186	0.1	0.074	0.062	0.131	0.13	0.155	0.089	0.124	0.109	0.105	0.132	0.117	0.129	0.07	0.152	0.129	0.185	0.156	0.123	0.163	0.06
190	0.065	0.037	0.078	0.08	0.041	0.038	0.051	0.088	0.057	0.09	0.092	0.058	0.066	0.052	0.043	0.371	0.074	0.165	0.069	0.129	0.14
192	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0.015	0	0
194	0.058	0.062	0.078	0.06	0.097	0.129	0.086	0.08	0.091	0.082	0.092	0.123	0.066	0.063	0.065	0.048	0.148	0.134	0.123	0.071	0.3
198	0.053	0.123	0.052	0.063	0.079	0.068	0.056	0.104	0.094	0.079	0.013	0.038	0.063	0.042	0.065	0.081	0.074	0.039	0.015	0.004	0
202	0.02	0.025	0.052	0.051	0.054	0.049	0.07	0.06	0.026	0.049	0	0.032	0.037	0.068	0.022	0	0.056	0.017	0.008	0.05	0.01
206	0.013	0	0.021	0.042	0.02	0.019	0.027	0.024	0.018	0.018	0.079	0.035	0.018	0.029	0	0	0.037	0.022	0.031	0.017	0
210	0.007	0.012	0.005	0.009	0.003	0	0.019	0.007	0.003	0.013	0	0.006	0.003	0	0	0	0	0	0	0	0
214	0	0.006	0.003	0	0	0	0	0.002	0	0.003	0	0.003	0.003	0	0	0	0	0	0	0	0
218	0.005	0.006	0.005	0.003	0	0	0.003	0	0.003	0	0	0	0	0	0	0	0	0	0	0	0
222	0	0	0	0	0.003	0	0	0	0	0	0	0	0.005	0	0	0	0	0	0	0	0
234	0	0	0	0	0	0	0	0	0.003	0	0	0	0	0	0	0	0	0	0	0	0
266	0	0	0	0	0	0	0	0	0	0	0	0	0	0.003	0	0	0	0	0	0	0
P-HW	0.146	<0.001	<0.001	0.855	0.778	0.595	0.532	0.140	0.808	0.920	0.166	0.125	0.351	0.347	0.635	0.514	0.981	0.023	0.912	0.031	0.834
Ots105																					
N	295	79	193	166	199	132	197	294	191	193	38	171	194	193	23	31	27	170	65	120	49
124	0	0	0	0.003	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
128	0.814	0.797	0.78	0.84	0.842	0.848	0.845	0.867	0.822	0.832	0.882	0.839	0.838	0.886	0.957	0.968	0.963	0.929	0.892	0.925	0.98
132	0.186	0.203	0.22	0.157	0.158	0.148	0.155	0.133	0.178	0.168	0.118	0.161	0.162	0.114	0.043	0.032	0.037	0.071	0.108	0.075	0.02
136	0	0	0	0	0	0.004	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
P-HW	0.705	0.725	1.000	0.629	0.792	1.000	1.000	0.020	0.805	0.440	0.414	0.154	0.791	1.000	1.000	1.000	1.000	0.582	0.143	0.124	1.000

Appendix 1. Continued.

Locus	Population																				
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21
Ots213																					
N	277	80	194	165	181	131	189	292	189	193	38	171	193	190	23	31	27	188	64	119	49
155	0.383	0.381	0.361	0.333	0.398	0.374	0.402	0.382	0.378	0.352	0.289	0.456	0.427	0.416	0.413	0.435	0.352	0.33	0.367	0.349	0.531
159	0.262	0.375	0.338	0.312	0.367	0.336	0.362	0.361	0.357	0.339	0.342	0.246	0.324	0.384	0.435	0.371	0.537	0.604	0.461	0.475	0.408
163	0.052	0.019	0.049	0.067	0.05	0.073	0.045	0.055	0.077	0.073	0.013	0.015	0.044	0.032	0.065	0.065	0.074	0.019	0.07	0.067	0.02
167	0	0	0.005	0	0	0	0	0	0	0.003	0	0	0	0	0	0	0	0	0	0	0
179	0	0	0	0	0	0	0	0	0	0.003	0	0	0	0	0	0	0	0	0	0	0
183	0	0	0	0	0	0	0	0	0	0	0.013	0	0	0	0	0	0	0	0	0	0
187	0	0	0.003	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
231	0.005	0	0	0.003	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
235	0.042	0.025	0.028	0.03	0.017	0.011	0.019	0.022	0.021	0.031	0.053	0.006	0.013	0.013	0	0.016	0	0	0	0	0
239	0.022	0.006	0.01	0.012	0	0	0.005	0.009	0.011	0.016	0.053	0	0.013	0.008	0	0	0	0	0	0.029	0.02
243	0.014	0.006	0.005	0.009	0.017	0.008	0.011	0.005	0	0.005	0	0	0.003	0.011	0	0	0	0	0	0	0
247	0.042	0.031	0.054	0.009	0.014	0.008	0.008	0.005	0.005	0.01	0	0.009	0.01	0.013	0	0	0	0.011	0.031	0.034	0
251	0.034	0.025	0.021	0.021	0.011	0.031	0.011	0.021	0.008	0.013	0	0.015	0.018	0.011	0	0	0	0	0	0	0
255	0	0	0.005	0.033	0.008	0.008	0.008	0.003	0.029	0.005	0	0.018	0.003	0	0	0	0	0.008	0.023	0.021	0
259	0.004	0.013	0.008	0.018	0	0.004	0.016	0.003	0.005	0.01	0.013	0.018	0	0.013	0	0	0	0	0	0	0
263	0.022	0	0.003	0.003	0.011	0	0	0.015	0.005	0.003	0	0.015	0	0	0	0	0	0.021	0.039	0.017	0
267	0.007	0.006	0.005	0.018	0.025	0	0.016	0.009	0.013	0.016	0.053	0.006	0.028	0.018	0	0	0	0	0	0	0
271	0.022	0.019	0.036	0.052	0.019	0.061	0.008	0.05	0.037	0.049	0.053	0.035	0.036	0.026	0.087	0.097	0.037	0.005	0	0	0
275	0.029	0.031	0.018	0.024	0.008	0.019	0.013	0.009	0.019	0.013	0	0.129	0.028	0.013	0	0	0	0	0	0.004	0.02
279	0.016	0.019	0.023	0.018	0.011	0.008	0.003	0.003	0	0.01	0.079	0.012	0.016	0.005	0	0.016	0	0	0	0	0
283	0.013	0	0.005	0.003	0.006	0.008	0.003	0.012	0.003	0	0	0.003	0.008	0	0	0	0	0	0	0	0
287	0.011	0.013	0.005	0	0	0.004	0	0.005	0.003	0.008	0	0.003	0	0.003	0	0	0	0	0	0	0
291	0.007	0	0.013	0.018	0.014	0.034	0.045	0.01	0.011	0.01	0.039	0.006	0.021	0.013	0	0	0	0	0.008	0	0
295	0.013	0.025	0.003	0.009	0.017	0.015	0.021	0.015	0.011	0.026	0	0	0.008	0.021	0	0	0	0.003	0	0.004	0
299	0	0.006	0	0.006	0.008	0	0.005	0.005	0.008	0.005	0	0.012	0	0	0	0	0	0	0	0	0
303	0.002	0	0.003	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0

Appendix 1. Continued.

													Population								
Locus	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21
Ots213																					
P-HW	0.442	0.246	0.509	0.075	0.120	0.656	0.078	0.127	0.025	0.216	0.452	0.680	0.730	<0.001	0.157	0.951	0.018	0.288	0.502	0.749	0.767
Ots2M																					
N	293	77	193	168	191	133	196	292	193	194	36	171	195	192	23	31	27	199	65	120	50
131	0.027	0	0.034	0.048	0.06	0.045	0.061	0.043	0.049	0.044	0.069	0.056	0.021	0.06	0.174	0.177	0.259	0.214	0.338	0.367	0.38
133	0.241	0.24	0.288	0.244	0.285	0.241	0.253	0.291	0.301	0.289	0.194	0.313	0.297	0.273	0.348	0.306	0.481	0.442	0.331	0.346	0.48
135	0.312	0.286	0.241	0.286	0.275	0.301	0.237	0.277	0.308	0.309	0.139	0.31	0.269	0.24	0.283	0.403	0.167	0.128	0.169	0.142	0.03
137	0.396	0.429	0.407	0.414	0.369	0.414	0.441	0.38	0.337	0.348	0.597	0.316	0.413	0.424	0.196	0.113	0.093	0.173	0.092	0.042	0.11
139	0.024	0.045	0.031	0.009	0.01	0	0.008	0.009	0.005	0.01	0	0.006	0	0	0	0	0	0.043	0.069	0.104	0
143	0	0	0	0	0	0	0	0	0	0	0	0	0	0.003	0	0	0	0	0	0	0
P-HW	0.850	0.289	0.380	0.027	0.526	0.263	0.468	0.677	0.135	0.414	0.620	0.167	0.829	0.018	0.300	0.614	0.502	0.062	0.051	0.126	0.400
Ssa407																					
N	277	80	193	168	193	136	184	291	192	188	38	171	186	193	23	31	27	184	65	120	50
149	0	0	0	0	0	0	0	0	0	0	0	0	0.003	0	0	0	0	0	0	0	0
153	0.144	0.163	0.127	0.149	0.14	0.158	0.174	0.16	0.174	0.136	0.079	0.237	0.183	0.233	0.152	0.242	0.204	0.082	0.069	0.15	0.04
157	0.023	0.019	0.023	0.015	0.01	0.015	0	0.002	0.01	0.005	0.013	0.006	0.003	0.023	0	0.016	0	0.024	0	0	0
161	0.029	0	0.005	0.006	0.008	0.004	0.005	0.021	0.008	0.005	0	0.009	0.016	0.003	0	0	0	0	0.008	0	0
165	0.15	0.106	0.114	0.101	0.163	0.088	0.12	0.115	0.099	0.12	0.184	0.111	0.126	0.075	0.196	0.016	0.037	0.03	0.031	0.017	0
169	0.307	0.356	0.363	0.366	0.28	0.294	0.293	0.323	0.281	0.335	0.289	0.196	0.309	0.272	0.065	0.145	0.074	0.049	0.146	0.158	0.17
173	0.128	0.169	0.155	0.095	0.135	0.173	0.111	0.143	0.143	0.128	0.171	0.102	0.102	0.132	0.065	0.113	0.056	0.079	0.169	0.158	0.11
177	0.005	0.013	0	0.027	0.023	0.037	0.03	0.014	0.026	0.013	0.013	0.079	0.005	0.01	0	0	0	0	0	0	0
181	0	0	0	0	0.003	0	0.003	0.007	0.003	0	0	0	0	0	0	0	0	0.003	0	0	0
185	0.005	0	0	0	0.003	0	0	0.003	0.008	0.011	0	0.012	0	0	0.022	0	0	0.068	0	0.004	0.02
189	0.029	0.037	0.036	0.03	0.034	0.029	0.024	0.015	0.018	0.024	0	0.003	0.024	0.01	0	0.032	0.019	0.109	0.146	0.046	0.03
193	0.067	0.031	0.054	0.051	0.065	0.081	0.054	0.06	0.083	0.04	0.026	0.05	0.073	0.052	0.022	0.016	0.037	0.117	0.085	0.142	0.08
197	0.038	0.056	0.062	0.06	0.07	0.066	0.106	0.072	0.065	0.061	0.171	0.094	0.105	0.101	0.283	0.242	0.241	0.231	0.215	0.196	0.38

Appendix 1. Continued.

Locus	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21
																		Population			
Ssa407																					
201	0.023	0.013	0.031	0.06	0.052	0.04	0.071	0.045	0.036	0.056	0.053	0.073	0.035	0.016	0.174	0.081	0.259	0.068	0.077	0.083	0.16
205	0.007	0	0.005	0.012	0.008	0.004	0.005	0.005	0.031	0.043	0	0.023	0.011	0.067	0.022	0.081	0.056	0.049	0.038	0.037	0.01
209	0.023	0.013	0.003	0.003	0	0	0	0.003	0	0	0	0	0	0	0	0	0	0.022	0	0	0
213	0.009	0.019	0.003	0.018	0.008	0.007	0.003	0.003	0.005	0.008	0	0.006	0.005	0.003	0	0.016	0.019	0.024	0.015	0.004	0
217	0.004	0.006	0.003	0.003	0	0.004	0	0.005	0.003	0.003	0	0	0	0.003	0	0	0	0.046	0	0.004	0
221	0.004	0	0.013	0.006	0	0	0	0.003	0	0.013	0	0	0	0	0	0	0	0	0	0	0
225	0.004	0	0.003	0	0	0	0	0	0.005	0	0	0	0	0	0	0	0	0	0	0	0
P-HW	0.483	0.141	0.609	0.295	0.472	0.446	0.471	<0.001	0.012	0.336	0.087	0.089	0.833	0.084	0.265	0.104	0.901	0.060	0.756	<0.001	0.092