

Sea Duck Joint Venture
Annual Project Summary for Endorsed Projects
FY04 – (October 1, 2003 to September 30, 2004)

Project Title: (SDJV #46) Population structure and annual survival estimation of female Black Scoters using genetic tagging

Principal Investigators: Sandra Talbot, Jason Schamber, Paul Flint, U.S. Geological Science Center, Alaska Science Center, 1011 E. Tudor Rd., Anchorage, Alaska, 99503; Matthew Perry, USGS, Patuxent Wildlife Research Center, 11410 American Holly Drive, Laurel, MD 20708-4015; Fred J. Broerman, USFWS, Yukon Delta NWR, PO Box 346, Bethel, Alaska 99559; sandy_talbot@usgs.gov

Project Description: Identify individuals through genetic techniques and estimate annual female survival using a mark-recapture approach. Use the same markers to lay the groundwork to examine population delineation in these species.

Objectives or Hypothesis:

1. Obtain an estimate of annual female survival based on collection and genetic analysis of contour feathers from nesting Black Scoter females.
2. Characterize population structure of Black Scoters at Aropuk Lake.

This project will fill the information gap and address high-priority needs identified by the Sea Duck Joint Venture Strategic Plan 2001 – 2006, by using population genetic analyses and genetic tagging in a mark-recapture study to define populations, estimate population size and trends, and understand population dynamics of a Black Scoter population.

Preliminary Results. We have obtained feather samples from over 30 nests from Aropuk Lake for each of three years (2002 – 2004). We have screened 70 microsatellite loci and typed 29 individuals from Aropuk Lake at 12 variable loci, to create a baseline dataset (data not shown). After informal exploration of data from these loci, we selected 8 (Table 1) for further consideration, based on published guidelines for choosing markers for genetic tagging studies (Paetkau 2000, 2003). Thus, we selected 8 loci with the highest mean expected heterozygosity (H_E), high resolution, with few or no 1-bp insertions, no initial indication of null alleles or allelic dropout, and relatively high variability.

After testing the loci for linkage disequilibrium and conformation to Hardy-Weinberg expectations, we used the statistical program, GIMLET 1.3.2 (Valieré 2003), to generate $P_{(IDObs)}$ and $P_{(IDSib)}$ values to determine whether the microsatellite markers are sufficiently variable to allow for use in individual identification. $P_{(IDObs)}$ is the probability at which another individual with the same genotype would be observed, given the sample frequency of the alleles observed at those loci, within the population. $P_{(IDSib)}$ estimates the probability of observing identical multilocus genotypes between two individuals sampled from a population comprised of first-order relatives (e.g., between siblings or parent-offspring). General guidelines for genetic

tagging studies suggest using a suite of markers that achieve a reasonably low $P_{(ID)}$ bounded between 0.01 and 0.0001; $P_{(IDSib)}$ provides a conservative upper bound on this estimate (Waits et al. 2001).

Table 1. Measures of genetic variability and probability of identity [$P_{(ID)}$] values for 8 microsatellite loci characterized from 29 Black Scoter females breeding at Aropuk Lake, Yukon Kuskokwim Delta, Alaska. Loci are ranked according to $P_{(ID)}$ value. $P_{(shadow)}$ is the probability that two individuals within the Aropuk Lake breeding population will share the same 8-locus genotype. H_E = expected heterozygosity; A = number of alleles.

Locus	H_E	A	$P_{(IDobs)}$	$P_{(IDSib)}$	Source
Aph07	0.89	12	0.0274	0.3185	Maak et al. 2000
Bcaμ11	0.79	9	0.0827	0.3809	Buchholtz et al. 1998
Aph04	0.68	4	0.1778	0.4593	Maak et al. 2003
CRG	0.65	3	0.2068	0.4821	A. Baker, unpublished
Aph18	0.63	4	0.2157	0.4944	Maak et al. 2003
Sfiμ11	0.51	2	0.3753	0.5942	Libants et al. 1999
Smo7	0.50	2	0.3837	0.6042	Paulus & Tiedemann 2003
Smo11	0.39	4	0.4064	0.6598	Paulus & Tiedemann 2003
Mean	0.63	5	-	-	
Multilocus product			1.052×10^{-6}	3.145×10^{-3}	
$P_{(shadow)}$			1/950,570	1/318	

We observed no significant linkage disequilibrium among the 8 loci tested. One locus, Aph18, demonstrated significant heterozygote deficit ($P < 0.018$). Probability of identity values indicate that an eight-locus genotype using these loci are sufficient to distinguish a single individual among 950,570 individuals drawn from the Aropuk Lake breeding population [$\text{multilocus } P_{(IDobs)} = 1.052 \times 10^{-6}$], and among 318 single-order relatives [$\text{multilocus } P_{(IDSib)} = 3.145 \times 10^{-3}$]. Since the Aropuk Lake population does not likely exceed 300 individuals (Schamber & Flint, unpublished data), a multilocus genotype comprised of these loci clearly has adequate statistical power for use in individual identification.

Although Aph18 is ranked 5th for resolving power, we will likely drop this locus from further analysis due to the finding of heterozygote deficit, a signal of possible genotyping error (Paetkau 2003). Analysis of data excluding Aph18 yields a multilocus $P_{(IDobs)}$ of 4.87×10^{-6} [$P_{(shadow)} = 1/205,339$] and a multilocus $P_{(IDSib)}$ of 6.36×10^{-3} [$P_{(shadow)} = 1/157$]. This 7-locus genotype is of lower resolving power but still adheres to guidelines recommending genetic tagging markers achieve a $P_{(ID)}$ bounded between 0.01 and 0.0001 (Waits et al. 2001).

Project Status. The work described above support both objectives of obtaining an estimate of female survival using genetic tagging for individual identification, and description of the population genetics characteristics of the Black Scoter population breeding at Aropuk Lake. We have not yet met the objective of comparing eastern and western populations; this will be accomplished upon analysis of samples collected from New Brunswick, Canada. Laboratory work is ongoing and we will proceed with multiplexing the microsatellite suite and analysis of contour feathers collected from 2002 – 2004. Laboratory analyses for the genetic tagging

portion of the study will be completed by December 31, 2004. Mark-recapture analysis will commence in January 2005. Genetic characterization of the Aropuk Lake and New Brunswick, Canada populations will be completed by August 30, 2005. A progress report will be submitted to the Sea Duck Joint Venture in May 2005, and a final report on October 1, 2005.

Project Funding Sources (US\$).

SDJV (USFWS)	Other U. S. federal contributions ¹	U. S. non-federal contributions	Canadian federal contributions	Canadian non-federal contributions	Source of funding (agency or organization)
\$16,129					
	\$28,530 ²				USGS/ASC
	\$ 5,000				USFWS/YDNWR
	not reported				USGS/Patuxent

¹values include matching funds.

²value includes matching funds estimated for field expenses (\$5000), laboratory expenses (\$4000) and Geneticists' salary associated with marker development and optimization (\$9000), and PI salary (\$10,530).

Total Expenditures by Category (US\$):

ACTIVITY	BREEDING ¹	MOLTING	MIGRATION	WINTERING	TOTAL
Banding					
Surveys					
Research	\$49,659				\$49,659
Communication					
Coordination					

¹value includes matching funds.

Literature Cited

Buchholtz W, Pearce JM, Pierson BJ, Scribner KT (1998) Dinucleotide repeat polymorphisms in waterfowl (family Anatidae): characterization of a sex-linked (z-specific) and 14 bi-parentally inherited loci. *Animal Genetics*, **29**, 323-325.

Libants S, Oswald K, Olle E, Scribner K (1999) Isolation of microsatellite loci from the spectacled eider: GenBank Accession Number AF180501. Available (September 28, 2004) at: <http://www.ncbi.nlm.nih.gov/entrez/viewer.fcgi?db=nucleotide&val=12734691>

Maak S, Neumann K, von Langerken G, Gattermann R (2000) First seven microsatellites developed for the Peking Duck (*Anas platyrhynchos*). *Animal Genetics*, **31**, 228-241

Maak S, Wimmers K, Weigend S, Neumann K (2003) Isolation and characterization of microsatellites in the Peking Duck (*Anas platyrhynchos*) and their application in other waterfowl studies. *Molecular Ecology Notes*, **3**, 224-227.

Paetkau D (2000) Genotyping errors in DNA-based inventories: a protocol that controls them, complete with empirical validation. Pages 12-13 *In*: Managing for Bears in Forested

Environments. Proceedings of the Columbia Mountains Institute of Applied Ecology Workshop, October 17-19, 2000, Revelstoke, British Columbia, Canada.

Paetkau D (2003) An empirical exploration of data quality in DNA-based population inventories. *Molecular Ecology*, **12**, 1375-1387.

Paulus KB, Tiedemann R (2003) Ten polymorphic autosomal microsatellite loci for the Eider duck *Somateria mollissima* and their cross-species applicability among waterfowl species (Anatidae). *Molecular Ecology Notes*, **3**, 250-252.

Valié N (2002) GIMLET: a computer program for analyzing genetic individual identification data. *Molecular Ecology Notes*, **2**, 377-379.

Waits LP, Luikart G, Taberlet P (2001) Estimating the probability of identity among genotypes in natural populations: cautions and guidelines. *Molecular Ecology*, **10**, 249-256.