

ALASKA BELUGA WHALE COMMITTEE
REPORT 05-2

**Biopsies of Bristol Bay Beluga Whales – A Genetic Mark-Recapture
Pilot Project, 2005**

Prepared by
Lori Quakenbush, Principal Investigator
Alaska Department of Fish & Game
1300 College Road
Fairbanks, AK 99701

A Cooperative Project among:

Bristol Bay Native Association

Ralph Andersen, Director, Natural Resources
Helen Chythlook, Coordinator, Special Projects
Hans Nicholson, Coordinator, Subsistence

Bristol Bay Marine Mammal Council

Myra Olsen, Chair
Nick Apokedak, Alternate Member and Beluga hunter

Alaska Beluga Whale Committee

Robert Suydam, Wildlife Biologist

National Marine Fisheries Service, Alaska Region Office

Barbara Mahoney, Biologist, Anchorage

Alaska Department of Fish and Game

Lori Quakenbush, Wildlife Biologist, Project Coordinator
Slim Morstad, Fisheries Biologist, Commercial Fisheries Division
Molly Chythlook, Subsistence Division

INTRODUCTION

Beluga whales (*Delphinapterus leucas*) are harvested for subsistence purposes in Bristol Bay. The Bristol Bay stock of beluga whales is one of five stocks that have been identified in Alaska based on their summering areas (Frost and Lowry 1990) and genetic structure (O’Corry-Crowe et al. 1997). From 1999 and 2000 aerial surveys, the population estimate for the Bristol Bay stock is approximately 2,000 belugas (Frost and Lowry 2002) and the stock appears to remain within the Nushagak and Kvichak drainages during the summer. Aerial surveys within the Kvichak in May and June 2002 and 2003 estimated 300–400 whales in the river system (Quakenbush 2003). We captured five whales in the Kvichak River in 2002 and 2003 as part of a cooperative study to determine the potential impact of beluga whales on a declining sockeye salmon (*Onchorynchus nerka*) population (Quakenbush 2003). At the 2003 Alaska Beluga Whale Committee annual meeting, scientists from the National Marine Fisheries Service, Marine Mammal Laboratory suggested applying genetics mark-recapture techniques to beluga stocks as a way to estimate stock size.

Genetic mark-recapture studies have been conducted using the uniqueness of an individual animal’s DNA as the mark (Palsbøll et al. 1997). A small skin sample provides enough genetic material for the analysis and it can be obtained from live animals using a biopsy dart. Marking the belugas would require getting skin biopsies that would be analyzed for individual DNA patterns. The recapture could come from harvested or stranded belugas that match the DNA from the original marks. Additional recaptures could be obtained from directed efforts to collect biopsies, which could also serve as additional marks. Biopsy techniques have been used to safely acquire samples from large cetaceans (Mathews et al. 1988, Whitehead et al. 1990, Brown et al. 1991, Barrett-Lennard et al. 1996, Hooker et al. 2001, Best et al. 2005), small cetaceans (Krützen et al. 2002), and pinnipeds (Gemmell and Majluf 1997, Wiig et al. 2000).

A critical component of a mark-recapture study is the number of marks in the population and the subsequent capture sample size. The number of marked animals required to achieve a given level of precision is related to the size of the population being sampled, the number of capture events, and the objectives of the study. We conducted a pilot study in 2004 to determine if beluga whales can be biopsied in numbers that justify a genetic mark-recapture study.

OBJECTIVES

1. Using the jabstick method developed in 2004 collect skin biopsies from up to 50 beluga whales in the Kvichak River to provide more genetic marks of the whales feeding there.
2. Travel to the Naknek River for reconnaissance purposes to plan for obtaining biopsies there in 2006.
3. Coordinate with beluga hunters to collect stomach contents from beluga whales harvested in the Kvichak River and analyze contents to determine species and amounts of prey consumed.
4. Analyze DNA in skin samples to determine if any belugas biopsied in 2005 were recaptures from 2004.

METHODS

Under National Marine Fisheries Service Permit # 782-1438 we were allowed to collect up to 50 biopsies from beluga whales to compare with the 30 obtained in 2004. We modified two 5-ft long wooden closet rails by drilling a threaded bolt in one end. The threaded end was also weighted with washers to allow the pole to land tip first. The threads allowed the biopsy tips to be screwed on and off. The end of the pole provided a penetration stop so that the biopsy was no deeper than the length of the tip (25 mm). The poles were made to be similar to harpoons used by native hunters. We tied the poles to the boat with a long line so that the pole could be thrown and pulled back into the boat.

We used one flat-bottom boat with a 40–60 hp, 4-stroke outboard and one 18-ft, aluminum Lund boat with a 70 hp, 2-stroke outboard motor. Local beluga hunters drove both boats familiar with herding individual whales or small groups of whales into shallow water. When whales began to touch the bottom, the boat driver would maneuver the boat to attempt to turn the whale. As the whale turned it would strand briefly allowing the boat to get close enough for the pole to be thrown or for the person with the pole to jab the whale. The boat would then retreat to allow the whale to move into deeper water.

For each whale biopsied, we recorded the number of whales in the group, the size and color of the whale, the size and color of any companions with the target whale, whether or not the companion whales was a calf, distance of the shot, reaction of the target whale to the biopsy and the reaction of other whales in the group, and the body location of the biopsy.

Sample numbers were written on waterproof paper, arranged sequentially, and placed in a waterproof bag. When a biopsy was taken, the tip was removed from pole and placed in a small whirlpak with one of the numbered labels. The number on the label was recorded on the data sheet to link the sample with the other observed information. Unused biopsy tips were stored in a Nalgene bottle in 70% ethyl alcohol so that a sterile tip could be quickly screwed on the jab stick for the next biopsy.

At the end of each day, the biopsy samples were removed from the tips and the skin was stored in a solution of 20% dimethyl sulfoxide (DMSO) saturated in salt (NaCl) in a labeled bottle. All used tips were washed with soap and water, dipped in bleach and placed in the Nalgene bottle of 70% ethyl alcohol. The skin will be transferred to the Southwest Fisheries Science Center in La Jolla, CA for genetic analysis. Our focus was on attaining skin samples; therefore we minimized the depth of the biopsy to 25 mm, which eliminated opportunities for blubber samples. If we had funding or specific objectives for blubber samples for contaminants or fatty acid analysis, we could have used longer biopsy tips and gotten blubber samples as well.

To minimize injury to belugas during this pilot study, we did not aim jabsticks at the head area, but considered any area posterior to the pectoral flippers and anterior to the peduncle as an acceptable target area. No jabstick was allowed to penetrate more than 25 mm. Belugas herded into shallow water for biopsy were pursued for no more than 10 minutes. If unsuccessful after 10 minutes, we moved to a different area to try again with different animals. We did not attempt to biopsy any new calves.

RESULTS

Using two boats and one jabstick per boat we were able to attain a total of 13 biopsies: four biopsies on 18 May, one on 19 May, and seven on 20 May (Table 1). All whales were within 3 m of the boat when biopsied. If the first attempt was missed, additional attempts were usually possible. Often the pole could be pulled back quickly and thrown again before the whale could move away from the boat. The mean number of attempts per sample was 2.5 (range 1–6). The most common reason for failure to get a sample was when the whale moved into deeper water. Another reason was the accuracy of the throw and the balance of the pole. Ideally the poles should be heavier overall and weighted to be front-heavy so that when thrown the tip hits the whale first.

Reactions to biopsy attempts were mostly related to whales attempting to avoid the approach of the boat than a reaction to the biopsy itself. The most common reactions we observed included acceleration and change of direction. We got most samples (54%) during high/slack and high/ebb tides (Table 1).

Calves accompanied three of the whales sampled. Overall, we were able to obtain 13 biopsies in three days with much more effort than last year due to the location of the whales much farther down river in more dynamic hydraulic conditions. Due to the difficulty we had in getting our desired sample size in the Kvichak River, we did not do a physical reconnaissance in the Naknek River, however we have made contacts with local experts there for assistance in 2006. No beluga hunting occurred while we were on the Kvichak, therefore no stomachs were collected.

DNA from the 13 biopsies collected in 2005 was compared with the 30 collected in 2004 and none of the individuals were recaptures between years. Two whales within each year however were sampled twice.

DISCUSSION

The beluga whales were much farther down the river between 18 and 20 May than previous years. We had to travel farther to find whales and had to work with them in areas where the hunters were less familiar. We suspect that the salmon smolt outmigration was early and that there were few smolt in the upper river for belugas to feed on by 18 May.

While we were not able to acquire the 50 samples that we needed for a mark-recapture study, we think that there may be an earlier time period worth trying. Belugas first come up the Kvichak River in April to feed on spawning rainbow smelt (*Osmerus mordax*) as the ice is breaking up. In 2005, hundreds of belugas were reported near Levelock, Alaska that were accessible for biopsies from shore (Nick Apokedak, pers. comm.). Sampling in April when the belugas are feeding on rainbow smelt and again during the salmon smolt outmigration in May, we might have two chances to attain the sample size we need.

Bristol Bay is a good location for a mark-recapture study because we have a reasonable idea of the overall population size. Local beluga hunters who know whale behavior and the

obstacles in the area are willing to participate. In addition to using the DNA for genetic mark-recapture studies, we can potentially acquire information about the relatedness of whales within a group, sex of individuals, site fidelity, movements, minimum ages of recaptured whales, and birth rates of individuals. In 2004, we collected samples from two adult whales that were traveling together and we may be able to determine their gender and relatedness.

CONCLUSIONS

1. Using a jabstick with a 25 mm biopsy tip and aluminum boats with outboard motors to herd beluga whales into shallow water was a safe and effective method of approaching belugas for skin samples.
2. Fewer samples were collected due to the location of the belugas farther downriver in areas less familiar to the boat drivers.
3. None of the samples collected in 2004 (n=11) were recaptures of those sampled in 2005 (n=28).

ACKNOWLEDGEMENTS

The success of this project has been due in large part to Nick and Brian Apokedak and Gusty Tallekpalek. Without their knowledge of beluga whale behavior and their boat driving skills we would not have been successful in getting skin samples. Their tolerance and patience in helping “gussaks” learn how to “harpoon” beluga whales was greatly appreciated. We thank Letty Hughes and Amy Frey for their assistance in the field. Helen Chythlook expertly recorded the data and helped with logistics. Rod Hobbs has been helpful with permit issues and determining necessary sample sizes. Greg O’Corry-Crowe and Amy Frey analyzed the skin samples to genetically identify the individual beluga whales. The Alaska Beluga Whale Committee funded the study and much personnel support and encouragement was provided by the Bristol Bay Native Association.

LITERATURE CITED

- Barrett-Lennard, L., T.G. Smith, and G.M. Ellis. 1996. A cetacean biopsy system using lightweight pneumatic darts, and its effect on the behavior of killer whales. *Marine Mammal Science* 12(1):14–27.
- Best, P.B., D. Reeb, M.B. Rew, P.J. Palsbøll, C. Schaeff, and A. Brandão. 2005. Biopsying southern right whales: their reactions and effects on reproduction. *Journal of Wildlife Management* 69(3):1171–1180.
- Brown, M., S.D. Kraus, and D.E. Gaskin. 1991. Reaction of North Atlantic right whales (*Eubalaena glacialis*) to skin biopsy sampling for genetic and pollutant analysis. Report of the International Whaling Commission, Special Issue 13:81–89.
- Frost, K.J., and L.F. Lowry. 1990. Distribution, abundance, and movements of beluga whales, *Delphinapterus leucas*, in coastal waters of western Alaska. Pages 39–57 in: T.G. Smith,

- D.J. St. Aubin, and J.R. Geraci, eds. Advances in research on the beluga whale, *Delphinapterus leucas*. Canadian Bulletin of Fisheries and Aquatic Sciences. Vo. 224.
- Frost, K.J., and L.F. Lowry. 2002. Alaska Beluga Whale Committee Survey of beluga whales in Bristol Bay, Alaska, 1999–2000. Alaska Beluga Whale Committee Draft Report 02–1. Pages 21–34 *in*: ABWC Final Report. NOAA #NA97FX0128.
- Gemmell, N.J. and P. Majluf. 1997. Projectile biopsy sampling of fur seals. *Marine Mammal Science* 13:512-516.
- Hooker, S.K., R.W. Baird, S. Al-Omari, S. Gowans, and H. Whitehead. 2001. Behavioral reactions of northern bottlenose whales (*Hyperoodon ampullatus*) to biopsy darting and tag attachment procedures.
- Krützen, M., L.M. Barré, L.M. Möller, M.R. Heithaus, C. Simms, and W.B. Sherwin. 2002. A biopsy system for small cetaceans: darting success and wound healing in *Tursiops* spp. *Marine Mammal Science* 18(4):863–878.
- Mathews, E.A., S. Keller, and D.B. Weiner. 1988. A method to collect and process skin biopsies for cell culture from free-ranging gray whales (*Eschrichtius robustus*). *Marine Mammal Science* 4(1):1–12.
- O’Corry-Crowe, G. M., R. S. Suydam, A. Rosenberg, K. J. Frost, and A. E. Dizon. 1997. Phylogeography, population structure and dispersal patterns of the beluga whale *Delphinapterus leucas* in the western Nearctic revealed by mitochondrial DNA. *Molecular Ecology* 6:955–970.
- Palsbøll, P.J., J. Allen, M. Bérubé, P.J. Clapham, T.P. Feddersen, P.S. Hammond, R.R. Hudson, H. Jørgensen, S. Katona, A.H. Larsen, F. Larsen, J. Lien, D.K. Mattila, J. Sigurjónsson, R. Sears, T. Smith, R. Spomer, P. Stevich, and N. Ølsen. 1997. Genetic tagging of humpback whales. *Nature*. Vol. 388:767–769.
- Quakenbush, L. 2003. Summer movements of beluga whales captured in the Kvichak River, in May 2002 and 2003. Alaska Beluga Whale Committee Report #03-03.
- Whitehead, H., J. Gordon, E.A. Mathews, and K.R. Richard. 1990. Obtaining skin samples from living sperm whales. *Marine Mammal Science* 6(4):316–326.
- Wiig, O., Berg, V., Gjertz, I., Seagars, D.J. and Skaare, J.U. 2000. Use of skin biopsies for assessing levels of organochlorines in walrus (*Odobenus rosmarus*). *Polar Biology* (2000) 23:272-278.

Table 1. Kvichak River beluga biopsy data, 2005.

Day in May	Time	Sample No.	Tide stage	Group size	Color of target	With a calf?	
						Yes	No
18	15:37	BB2005-01	high/ebb	2	gray		No
18	15:50	BB2005-02*	high/ebb	1	white		No
18	16:00	BB2005-03*	high/ebb	2	white		Yes
18	16:20	BB2005-26	high/ebb	1	white		No
19	15:50	BB2005-04	high/ebb	2	white		Yes
19	17:23	BB2005-27	high/ebb	2	white		Yes
20	11:40	BB2005-28	low/slack	4	white		No
20	12:30	BB2005-05	low/flow	1	white		No
20	12:35	BB2005-06	low/flow	2	white		Yes
20	13:10	BB2005-07	med/flow	4	white		No
20	13:20	BB2005-08**	med/flow	2	gray		Unkown
20	13:35	BB2005-09**	high/slack	6	gray		No
20	15:25	BB2005-10	med/ebb	6	white		No

* These two samples are from the same individual

** These two samples are from the same individual

