ALASKA BELUGA WHALE COMMITTEE
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Biopsies of Bristol Bay Beluga Whales – A Genetic Mark-Recapture Pilot Project

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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, Dr. Sue Moore of the Marine Mammal Lab, National Marine Fisheries Service suggested that genetics mark-recapture techniques applied to beluga stocks might be 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), 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 sample size. The number of animals that need to be marked and recaptured is related to the overall population being sampled, the original number that can be marked, and the number that can be sampled for recaptures. We conducted a pilot study to determine if beluga whales can be biopsied in numbers that justify a genetic mark-recapture study.

OBJECTIVES

1. Test different biopsy methods to determine, which is the safest and most effective for the Bristol Bay area and for beluga whales in general.
2. Test different methods of approaching beluga whales to determine the safest and most effective for the Bristol Bay area and for beluga whales in general.
3. Collect skin biopsies from up to 30 beluga whales in the Kvichak and Naknek rivers to determine if a genetic mark-recapture study is feasible in Bristol Bay.

METHODS

Sample size

We applied to the National Marine Fisheries Service for an increase in the number of biopsies allowed for the Bristol Bay stock allowed under Scientific Permit # 782-1438. We determined that biopsies from 30 belugas would allow us to test whether a genetic mark-recapture study was viable.

Biopsy Equipment Tested

Crossbow. We tested standard crossbow arrows made buoyant with foam collars and fitted with biopsy tips (manufactured by Finn Larsen, 2.5 cm-long x 0.6 cm diameter fitted with dental barbs to hold sample material). The arrows were shot from crossbows (Barnett WildCat with 150 lb prod) at foam targets on the beach at various distances. We also tested buoyant arrows on floating targets and tethered arrows on both targets. We did not try biopsy darts from modified rifles or shotguns as we did not want to reduce our chances for additional biopsies by disturbing the groups with the associated noise.

Jabstick. We modified two 4-ft long poles by drilling a threaded bolt in one end. 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.

Two 18-ft aluminum Lund boats (Nick's boat with a 70 hp motor and Gusty's boat with an 30 hp motor) driven by local beluga hunters were used to herd 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 crossbows or 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 dart or 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

Crossbow

We sighted-in the crossbows at 15 m using the buoyant arrows and practiced for proficiency. Tethered arrows were not reliable. The retrieval line easily tangled and did not play out well. The arrow also dropped more quickly than the buoyant arrows. We also practiced on floating targets and found that if the tip of the arrow hit the water, the arrow deflected due to the buoyant collar on the arrow. Therefore, the shooter could only target the area above the waterline.

We attempted to position the boat alongside swimming belugas for a chance with the crossbow. Belugas swimming near the boat did not present a very large target as only some of the head and back was exposed upon surfacing. Shooting at a relatively small moving target from a moving platform proved to be problematic and no belugas were biopsied using this method.

We tried a location on shore of an island (Gilligan's) in the Kvichak River that had a channel that ran near the island's shore. We positioned two people with crossbows on the shore and waited as the tide went out. Three whales came within range. Four attempts were made but none were successful. We also tried to move the whales closer to the shooters by placing a boat on the opposite side of the channel without success.

Jabstick

Using two boats and two jabsticks we were able to attain 17 biopsies on 21 May, 10 on 22 May and the remaining three that we were allowed by permit on 23 May (Table 1). Gusty's boat was responsible for 13 of the 30 samples and Nick's boat was responsible for 17 samples.

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 weighted to be front-heavy so that when thrown the tip hits the whale first. Our poles were not weighted properly and were more effective for jabbing then throwing.

Reactions to biopsy attempts were more 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 (43%) during high/slack tides (Table 1). We got fewer samples at other tide stages, however the important factor seemed to be the availability of shallow water.

Six of the whales sampled were accompanied by calves, three of which were new calves. In one instance, we were able to sample two white whales traveling together. Overall, we were able to obtain 30 biopsies in less than three days and believe that 30 more would have easily been possible.

DISCUSSION

Bristol Bay is a good place 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 this endeavor.

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. We collected samples from two adult whales that were traveling together and will be able to determine their gender and relatedness.

Although we were successful in attaining our objectives it will be possible to reduce our number of attempts per sample with front-weighted poles. Weighting the jabstick so that the tip drops quickly will allow the tip to hit the whale first. A jabstick that is not weighted tends to drop parallel to the whale instead of closer to perpendicular.

CONCLUSIONS

1. Using a jabstick with a 25 mm biopsy tip was the safest and most effective method of obtaining skin samples.
2. Using aluminum boats with outboard motors to herd beluga whales into shallow water was the safest and most effective method of approaching belugas for skin samples.

3. We obtained 30 skin samples in less than three days and believe it would be possible to get 60 in five or six days.

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Table 1. Kvichak River beluga biopsy data, 2004.

Sample No.	Day in May	Time	Tide stage	Group size	Color of target	With a calf? Yes	No
BB2004-11	21	13:25	low/ebb	1	white-gray		No
BB2004-01	21	13:45	low/slack	1	gray		No
BB2004-02	21	15:30	low/flow	5	white	Yes (new)	
BB2004-12	21	17:45	high/slack	2	white	Yes	
BB2004-03	21	17:50	high/slack	1	white		No
BB2004-13	21	17:55	high/slack	2	white-gray	Yes (new)	
BB2004-14	21	18:15	high/slack	2	white		No
BB2004-15	21	18:18	high/slack	2	white		No
BB2004-04	21	18:35	high/slack	2	white		No
BB2004-05	21	18:55	high/slack	2	white	Yes (new)	
BB2004-06	21	19:10	high/slack	2	white		No
BB2004-16	21	19:15	high/slack	8	white		No
BB2004-07	21	19:18	high/slack	2	gray		No
BB2004-17	21	19:50	high/ebb	2	white-gray	Yes	
BB2004-08	21	20:00	high/ebb	5	white		No
BB2004-18	21	20:00	high/ebb	1	white		No
BB2004-09	21	20:08	high/ebb	2	white-gray		No
BB2004-19	22	14:25	low/slack	2	white-gray		No
BB2004-20	22	14:35	low/slack	5	white-gray		No
BB2004-10	22	14:40	low/slack	5	gray		No
BB2004-21	22	15:10	low/slack	1	gray		No
BB2004-22	22	15:50	low/flow	1	white		No
BB2004-26	22	16:00	low/flow	10	gray		No
BB2004-23	22	16:05	low/flow	1	white-gray		No
BB2004-24	22	19:45	high/slack	2	gray		No
BB2004-25	22	20:00	high/slack	2	white	Yes	
BB2004-27	22	20:00	high/slack	6	gray		No
BB2004-29	23	12:18	high/ebb	2	white		No
BB2004-30	23	13:35	low/ebb	50	gray		No
BB2004-28	23	13:40	low/ebb	50	gray		No