Behavioral reactions of northern bottlenose whales (*Hyperoodon ampullatus*) to biopsy darting and tag attachment procedures

Sascha K. Hooker
Robin W. Baird
Sa'ad Al-Omari
Shannon Gowans
Hal Whitehead

Biology Department
Dalhousie University
Halifax, Nova Scotia
B3H 4J1, Canada
Present address (for S. K. Hooker): British Antarctic Survey
High Cross, Madingley Road
Cambridge, CBE OET, United Kingdom
E-mail address (for S. K. Hooker):skh@bas.ac.uk

Abstract—The effects of invasive or intrusive research techniques need to be thoroughly documented in order to satisfy appropriate standards of animal care. How cetaceans react to either biopsy darting or tag attachment procedures has been studied for several species, and considerable interspecific variability in responses has been demonstrated; however, few studies have compared reactions to both techniques. In the family Ziphiidae (the beaked whales) nothing has been previously reported on responses to either technique. We examined and compared the reactions of northern bottlenose whales (*Hyperoodon ampullatus*) to biopsy darting and tagging. Reactions to both these procedures were generally low-level and short-lived; stronger responses were given to hits than to misses. There was no statistical difference in observed response to tag versus biopsy hits. The prior behavioral state of the whales appeared to influence the magnitude of reaction to both hits and misses and thus may be an important factor to consider in such impact assessment. Whales lying still at the surface showed stronger reactions than traveling or milling animals. Sea state appeared to affect whether there was a reaction to misses. Whales were more likely to respond to a miss in calm sea conditions. No avoidance of the research vessel was observed following a tag or biopsy attempt, and in most cases whales approached the research vessel again within several minutes.

Manuscript accepted 16 November 2000.
Fish. Bull. 99:303–308 (2001).

The nonlethal firing of projectiles at whales and dolphins is increasingly being used, both in order to obtain skin and blubber samples (e.g. Lambertsen, 1987) and to attach data-recording or transmitting devices (e.g. Mate and Harvey, 1983; Goodyear, 1993; Baird, 1998; Mate et al., 1998). Data collected with these techniques are important for management and conservation purposes but may come at some cost (usually a behavioral disturbance) to the individuals involved. This cost may vary for different species or populations (see e.g. Schneider et al., 1998), therefore the impacts should be assessed each time a study is conducted.

Reactions of various species of cetaceans to biopsy darting have generally been mild (e.g. International Whaling Commission, 1989; Whitehead et al., 1990; Brown et al., 1991; Weinrich et al., 1991, 1992; Barrett-Lennard et al., 1996; Weller et al., 1997). The most common response is a "startle" reaction, although the level of reaction varies slightly between species, and also between populations and individuals. In contrast, the reaction of cetaceans to tagging with suction-cup–attached tags has been found to vary dramatically. Although reactions of killer whales (*Orcinus orca*) and Dall's porpoises (*Phocoenoides dalli*) to the technique were minor (Baird, 1994; Hanson and Baird, 1998), those of bottlenose dolphins (*Tursiops* sp.) were strong and relatively long-lasting (Schneider et al., 1998).

The family Ziphiidae (the beaked whales) is the second largest family of cetaceans, yet no studies have reported their reactions to tagging or biopsy techniques. In this paper we compare the reactions of northern bottlenose whales (*Hyperoodon ampullatus*) to both techniques and investigate factors affecting the behavioral reactions observed. These results are particularly relevant to agencies that grant research permits (e.g. the National Marine Fisheries Service in the USA), which often require some discussion of the implications of research techniques in terms of animal care. Additionally, assessment of the magnitude and duration of any behavioral response caused by the process of attaching a tag is vital in ensuring that the attachment of the tag does not confound the behavioral data it records.

Materials and methods

Field research took place off eastern Canada, approximately 300 km east of Halifax, Nova Scotia, over a submarine canyon termed the "Gully" (approximate position: 44°N, 59°W) during June–August 1996–98. All tagging or biopsy attempts were made opportunis-

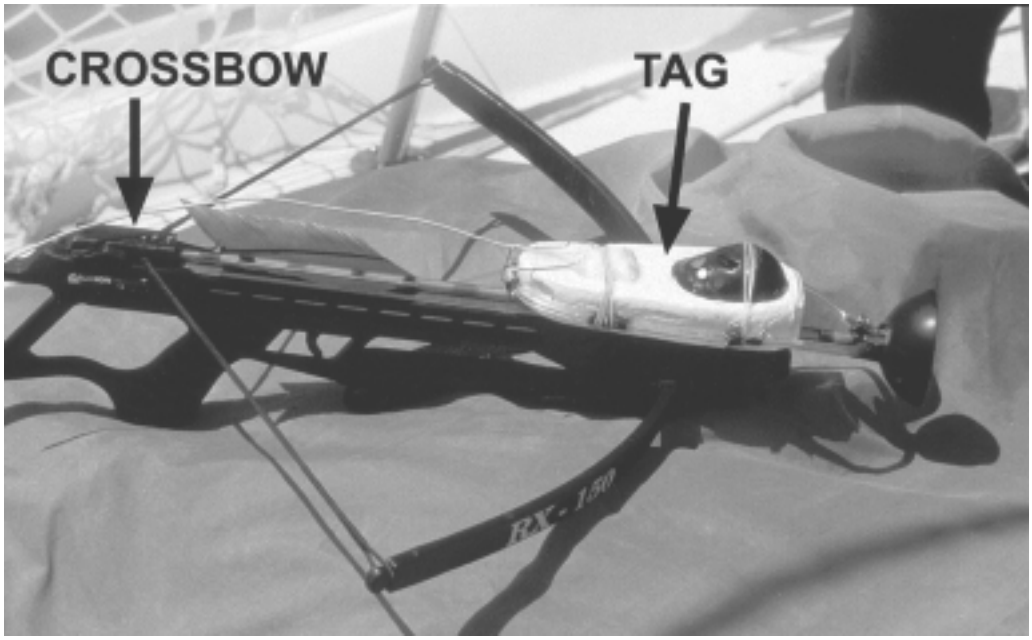

Figure 1

Photograph of the suction-cup–attached time-depth recorder and VHF radio tag ready to be deployed from a crossbow.

tically from a 12-m sailing vessel, operated under power at speeds of 1–4 knots.

The biopsy dart had a 2.5-cm-long, 0.6-cm-diameter cylindrical punch fitted with a dental broach (a barbed filament to hold a sample in place) (as illustrated in Barrett-Lennard et al., 1996), attached to the end of a standard crossbow bolt (total weight 28.5 g). A cylindrical stopper, set 2.5 cm back from the tip of the punch, caused the bolt to rebound after impact with the whale. Bolts were fired from a 67-kg-draw crossbow (Barnett WildCat XL) at a range of 5–15 m. Samples were usually taken from the flank near the dorsal fin. The floating dart was recovered and the skin and blubber sample was removed. The tissue was then subsampled for various analyses requiring either skin or blubber or both. The gender of the biopsied whales in this study was determined genetically (Gowans et al., 2000).

The tag measured 20 · 4 · 5 cm and had a 40-cm flexible antenna. The foam housing of the tag contained a time-depth recorder (Wildlife Computers, Redmond, WA, or AGO Environmental Electronics, Victoria, BC) and a VHF radiotransmitter (Advanced Telemetry Systems, Isanti, MN, or Telonics, Mesa, AZ). An 8-cm-diameter rubber suction cup (designed for automobile roof-racks, Canadian Tire) was used to attach the tag. The total unit weighed approximately 340 g in air. Tags were attached to a modified crossbow bolt (weight approximately 25 g) and were deployed with the same crossbow as described above (Fig. 1).

The group size and behavioral state of the whales prior to the biopsy darting or tagging attempt were noted. Group size was defined as the number of animals at the surface within five body lengths of each other (chain rule, see Smolker et al., 1992). Behavioral state was assigned as either logging (lying still or moving slowly in one direction at the surface) or milling or traveling (milling—moving slowly in no consistent direction; traveling—moving in a consistent direction at greater than 2 knots). Whenever possible, each tag or biopsy attempt was videotaped and this recording was used to confirm the consistency of behavioral categories assigned by different observers in the field for both before tagging attempt behavior and reaction. Attempts were classified as a hit or a miss; a hit was defined as contact with the whale and hits were further subdivided as to whether they were successful, i.e. whether biopsies obtained a sample or whether tags remained attached to the whale for more than 30 seconds. Sea state (Beaufort scale) was recorded every hour; sea state at the time of the biopsy or tag attempt was interpolated from these hourly logs. Categories of reaction types were defined following Weinrich et al. (1991):

1 No reaction: whale continued to show the same behavior as before the biopsy or tagging attempt;
2 Low-level reaction: whale modified its behavior slightly, e.g. dived rapidly or flinched;
3 Moderate reaction: whale modified its behavior in a more forceful manner but gave no prolonged evidence of behavioral disturbance, e.g. tail slap, acceleration, and rapid dive;
4 Strong reaction: whale modified its behavior in a succession of forceful activities, e.g. successive percussive behaviors (breaches, tail slaps).

Table 1

Number of whales showing different reaction-types to tagging and biopsy deployment attempts (percentages shown in parentheses).

	Reaction level			
Event	None	Low-level	Moderate	Total no. of whales
Tag-hit	2 (7%)	19 (65%)	8 (28%)	29
Biopsy-hit	3 (11%)	20 (74%)	4 (15%)	27
Tag-miss	33 (60%)	21 (38%)	1 (2%)	55
Biopsy-miss	16 (80%)	3 (15%)	1 (5%)	20

Goodness-of-fit *G*-tests were used to compare reactions to different techniques and under different conditions. Small sample sizes often necessitated pooling between categories. Because reactions to misses tended to be lower-level they were pooled between none and low to moderate, whereas reactions to hits tended to be higher-level and were pooled between none (or low) and moderate.

Results

Forty-seven biopsy attempts were made on northern bottlenose whales in 1996 and 1997 (Table 1). Of these, 27 attempts hit the whale and 20 obtained a skin and blubber sample. Six attempts hit, but did not retain a sample (primarily resulting from low hits at the water line). One dart sank after hitting a whale. Video footage was taken of 18 biopsy hits (15 successful) and 11 biopsy misses.

Eighty-four tagging attempts were made between 1996 and 1998 (Table 1). Twenty-nine attempts hit the whale but only six successfully attached (one for only 30 seconds). Video footage was taken of 15 tag hits (three successful attachments) and 34 tag misses.

The majority of whales showed no or low-level reactions to both tag and biopsy attempts (Table 1). No strong reactions were observed during our study. The whales reacted to 93% of the tag hits and 89% of the biopsy hits, but reacted to only 40% of the tag misses and 20% of the biopsy misses (Table 1). Reactions (categorized as none, low, or moderate) were significantly greater for hits than for misses (G=50.3, P<0.001, 2 df). Reactions to tag hits were not significantly different from the reactions to biopsy hits (G=1.51, P=0.47, 2 df). Of the 6 successful tag hits, 2 animals gave a hard tail flick, accelerated and dove (moderate reaction), 1 animal accelerated and dove, and 3 dove rapidly or flinched (low-level reactions).

The type of reaction to a hit was significantly related to the animal's behavioral state prior to the tagging or biopsy attempt (comparison for all hits of behavioral state (logging vs. milling or traveling) and reaction (none [or low] vs. moderate) G=4.04, P=0.044, 1 df). Low-level reactions were most common for traveling or milling whales, whereas logging whales were more likely to show stronger reactions, especially to tag hits (Fig. 2). There was also a similar relationship between behavior and reaction to misses, although whales rarely showed a moderate reaction to a miss (comparison of behavioral state [as above] and reaction [none vs. low or moderate] G=4.02, P=0.045, 1 df, Fig. 2). Reactions to hits of animals in groups were similar to those for lone individuals (G=0.767, P=0.38, 1 df). There was an effect of sea state on reaction type, but only for missed shots (comparison of sea state [≥Beaufort 2 with Beaufort <2], G=4.38, P=0.036 1 df, Fig. 3). This effect was greater for tag misses (G=6.80, P=0.009, 1 df) than for biopsy misses (G=2.41, P=0.12, 1 df).

Table 2

Gender of biopsied whales (n=20) and respective reactions.

	Reaction level			
Gender	None	Low-level	Moderate	Total no. of whales
Male	0	5	2	7
Female	2	9	2	13

No avoidance of the research vessel was observed after tagging or biopsy attempts. For the majority of attempts, animals remained at the surface. If the animals made a shallow dive, they usually returned to the research vessel within a few seconds or were photographed again within a few minutes of the tagging or biopsy attempt (for 82% of all attempts, whales returned to the surface and remained with the research vessel for at least five minutes). There was little difference in the likelihood of encounters ending within five minutes between tagging and biopsy attempts or between hits and misses. Furthermore, encounters including tagging or biopsy attempts were not shorter in duration than encounters without any tagging or biopsy attempt (one-tailed t-tests, P>0.05).

No significant difference between the reactions of males and females was found (G=2.1, P=0.35, 2 df, Table 2), but samples sizes were small and did not adequately represent the population.

Discussion

The response rate of northern bottlenose whales to biopsy hits (89%) was greater than that found for baleen whales (right whales, *Eubalaena glacialis*, 19%; Brown et al., 1991; humpback whales, *Megaptera novaeangliae*, 50%, Weinrich et al., 1991), but was similar to that recorded for other odontocetes. A 100% response rate was found for sperm whales (*Physeter macrocephalus)* (Whitehead et al., 1990) and for bottlenose dolphins (Weller et al., 1997), and an 81% response rate was found for killer whales (Barrett-Lennard et al., 1996). Reactions of bottlenose dolphins consisted of an observable short-term change in behavior (Weller et al., 1997). Momentary shakes or accelerations were observed for killer whales (Barrett-Lennard et al.,

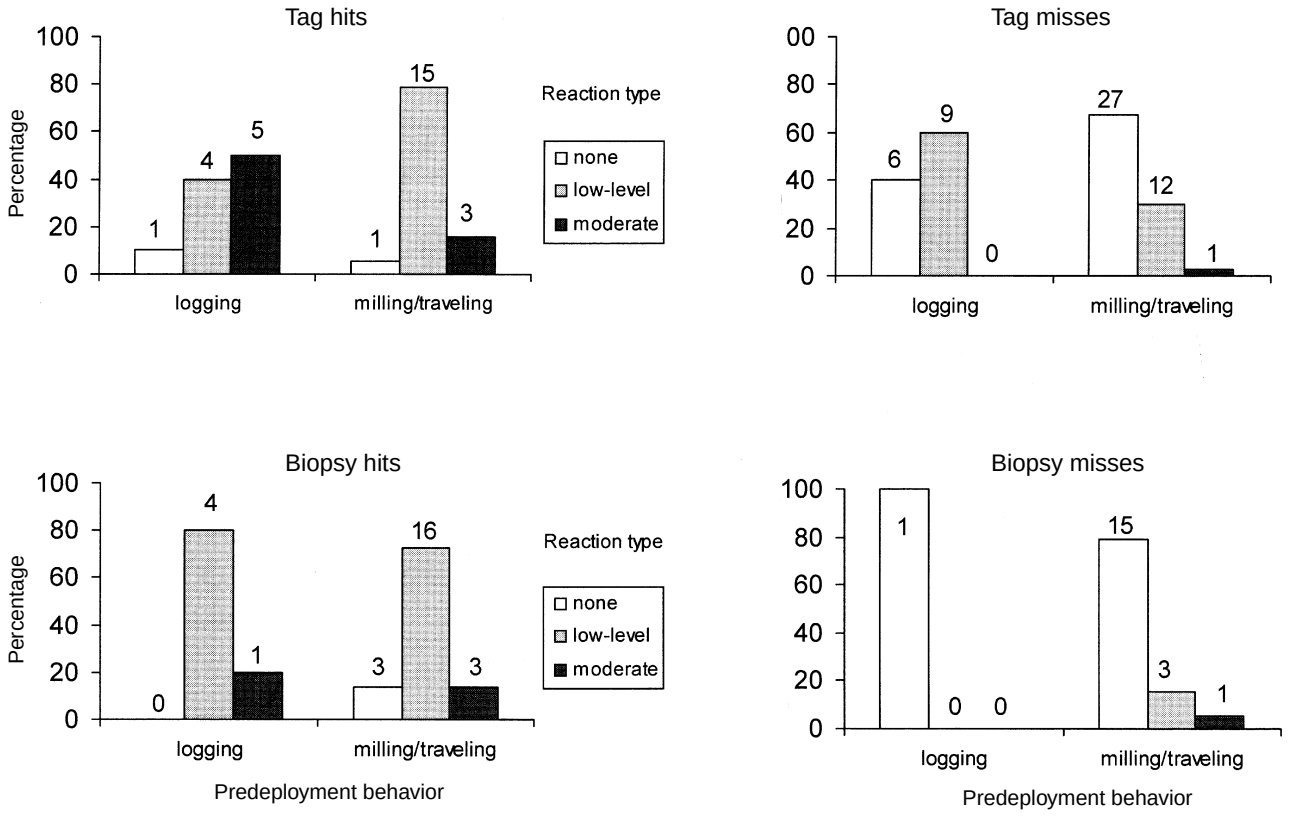

Figure 2

Relationship between behavioral state and reaction type for tag and biopsy deployments. Reactions during logging behavior were stronger than for other behaviors for both hits (P=0.044) and misses (P=0.045). Percentage of each reaction-type is displayed for each prebiopsy behavioral category (number of reactions of each type are shown above each bar).

1996), whereas sperm whales showed strong startle reactions, occasionally involving defecation (Whitehead et al., 1990). Reactions of bottlenose whales seemed most similar to killer whale reactions, i.e. relatively "low-level." Gender-related differences in the levels of reaction have previously been noted for humpback whales (Brown et al., 1994), although no such effects were found in this study.

Reactions of northern bottlenose whales to suction-cup tag deployment were also low-level. Bottlenose whales reacted to 93% of tag hits and 40% of tag misses. Baird (1994) documented only minor reactions by killer whales to crossbow-deployed suction-cup–attached tags: 52% reaction to hits and 26% reactions to misses. Reactions of short-finned pilot whales (*Globicephala macrorhynchus*) to the same tags deployed by crossbow consisted of a tail flick and rapid dive (Baird[1]). Similar variation in reaction has been observed from suction-cup–attached tags deployed by pole. Little reaction was observed to tags deployed on Dall's porpoise (Hanson and Baird, 1998) or pantropical spotted dolphins (*Stenella attenuata*; Baird[1]), whereas bottlenose dolphins showed prolonged and intense reactions to tagging attempts, reacting to 100% of hits and 71% of misses (Schneider et al., 1998). The scale of reaction for northern bottlenose whales appears to lie in the middle of this range.

As noted, reactions to tag hits were similar to reactions to biopsy hits, despite the fact that tags weigh substantially more than biopsy darts. A potential explanation for this is the variation in "tag hit" under our definition: some of the tag hits were glancing blows and so may have had little striking impact, whereas others were direct hits. In contrast, the impact of biopsy darting was more consistent (i.e. there were virtually no glancing hits for biopsy darts). In addition, the greater weight of the tags resulted in a slower delivery speed, thus the force of a heavier, slower-moving tag may have been similar to that of the lighter, faster-moving biopsy dart. Alternatively, animals may have a set reaction to any impact, such that the same response will be elicited unless some threshold is exceeded (perhaps through excessive or repeated impact).

As many authors have pointed out, evaluating short and long-term reaction to tag attachment is important in determining whether the presence of tags affects the recorded behavior (White and Garrott, 1990; Walker and Boveng, 1995; Croll et al., 1996). Data were recovered from two of

[1] Baird, R.W. 1999. Unpubl. data. Biology Department, Dalhousie University, Halifax, NS, B3H 4J1 Canada.

the six successful tag attachments and one of these tags was equipped to record velocity (Hooker and Baird, 1999). The whale's initial reaction to the attachment of the tag was a rapid acceleration and dive. The velocity record showed that this initial increase dropped within the first two minutes to levels observed for the rest of the deployment (Hooker and Baird, 1999). The general behavior of all six tagged whales (in terms of surfacing intervals and dive durations) was also consistent with that observed from nontagged whales. Thus, although based on a small sample size, it appears that the target animals' behavior is modified for only a few minutes in a short-term reaction to the tagging procedure.

Previous studies that examined responses to biopsy darting have been criticized because of the potentially confounding effect of the research vessel approach (Brown et al., 1994). In our study, the behaviors of the whales when first sighted did not change in any noticeable way during the approach of the research vessel for photo-identification, and immediately prior to the tagging or biopsy attempt. Thus we are relatively confident that the reactions we observed were due to the tagging or biopsy darting, rather than to the proximity of the research vessel. However, the approach of the research vessel may have caused subtle changes in behavior that we did not observe.

The major cue to which bottlenose whales react appears to be the physical impact of the tag or biopsy, because reaction to hits was much greater than reaction to misses (Table 1). Whales also appear to react to an acoustic or other sensory cue, evidenced by their stronger reactions to tag misses in calm sea conditions (when the hit would be more audible or the splash more likely to be detected above background levels; Fig. 3).

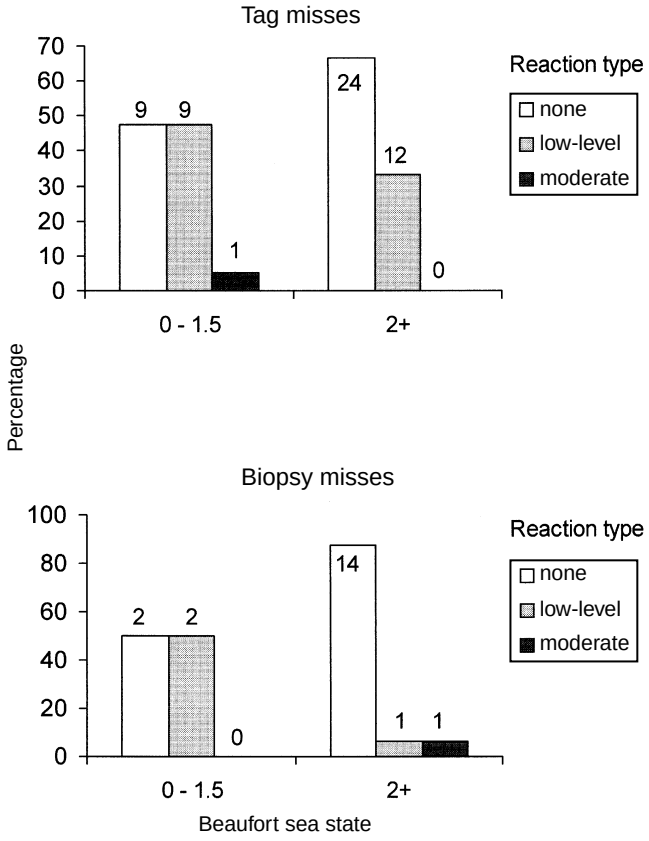

Figure 3

Relationship between sea state and reaction type for tag and biopsy misses. Reactions to misses were greater in calm (<Beaufort 2) than in rough (≥Beaufort 2) sea states (P=0.036). Percentage of each reaction-type is displayed for these two Beaufort sea state categories (number of reactions of each type are shown above each bar).

The primary factor affecting the reaction of bottlenose whales to either tag or biopsy stimulus appears to be the behavior of the whale at the time of the stimulus. The relative stimulus of a biopsy or tag is less for whales involved in traveling or milling than for whales lying still at the surface (Fig. 2). Similarly, differences in the level of reaction have been observed for humpback whales involved in different activities. Reactions of migrating humpback whales were generally lower than those of whales on feeding or breeding grounds (Brown et al., 1994), and mothers or primary escorts of humpback whales (thought to be involved in breeding activity) showed less reaction than other whales on the breeding grounds (Clapham and Mattila, 1993). Such results have wide implications for monitoring the effect of various activities (e.g. noise pollution) on cetaceans because the likelihood of reaction may vary depending on behavioral state.

Acknowledgments

We are grateful to all the crew members of *Balaena* who participated in fieldwork in the Gully. Research in the Gully was funded by the Natural Sciences and Engineering Research Council (NSERC), World Wildlife Fund Canada, and the Canadian Federation of Humane Societies. Merel Dalebout (University of Auckland) determined the gender of biopsied whales. S.K.H. was supported by a Canadian Commonwealth Scholarship, R.W.B. by an NSERC postdoctoral fellowship, and S.G. by an NSERC and an Izaak Walton Killam Memorial Scholarship. Lisa Balance, David Maehr, and Per Palsboll made helpful comments on the manuscript.

Literature cited

Baird, R. W.
1994. Foraging behaviour and ecology of *transient* killer whales (*Orcinus orca*). Ph.D. diss., Simon Fraser Univ., Burnaby, British Columbia, 157 p.
1998. Studying diving behavior of whales and dolphins using suction-cup attached tags. Whalewatcher: J. Am. Cetacean Soc. 31(1):3–7.

Barrett-Lennard, L. G., 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. Mar. Mamm. Sci. 12:14–27.

Brown, M. R., P. J. Corkeron, P. T. Hale, K. W. Schultz, and M. M. Bryden.
1994. Behavioral responses of east Australian humpback whales *Megaptera novaeangliae* to biopsy sampling. Mar. Mamm. Sci. 10:391–400.

Brown, M. W., 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. Rep. Int. Whal. Comm. Spec. Issue 13:81–89.

Clapham, P. J., and D. K. Mattila.
1993. Reactions of humpback whales to skin biopsy sampling on a West Indies breeding ground. Mar. Mamm. Sci. 9:382–391.

Croll, D. A., J. K. Jansen, M. E. Goebel, P. L. Boveng, and J. L. Bengtson.
1996. Foraging behavior and reproductive success in chinstrap penguins: the effects of transmitter attachment. J. Field Ornith. 67:1–9.

Goodyear, J. D.
1993. A sonic/radio tag for monitoring dive depths and underwater movements of whales. J. Wildl. Manage. 57: 503–513.

Gowans, S., M. L. Dalebout, S K. Hooker, H. Whitehead.
2000. Reliability of photographic and molecular techniques for sexing northern bottlenose whales (*Hyperoodon ampullatus*). Can. J. Zool. 78:1224–1229.

Hanson, M. B., and R. W. Baird.
1998. Dall's porpoise reactions to tagging attempts using a remotely-deployed suction-cup tag. Mar. Tech. Soc. J. 32(2): 18–23.

Hooker, S. K., and R. W. Baird.
1999. Deep-diving behaviour of the northern bottlenose whale, *Hyperoodon ampullatus* (Cetacea: Ziphiidae). Proc. Roy. Soc. London B 266:671–676.

International Whaling Commission.
1989. Report of the sub-committee on small cetaceans. Rep. Int. Whal. Comm. 39:117–129.

Lambertsen, R. H.
1987. A biopsy system for large whales and its use for cytogenetics. J. Mammal. 68:443–445.

Mate, B. R., and J. T. Harvey.
1983. A new attachment device for radio-tagging large whales. J. Wildl. Manage. 47:868–872.

Mate, B. R., R. Gisiner, and J. Mobley.
1998. Local and migratory movements of Hawaiian humpback whales tracked by satellite telemetry. Can. J. Zool. 76:863–868.

Schneider, K., R. W. Baird, S. Dawson, I. Visser, and S. Childerhouse.
1998. Reactions of bottlenose dolphins to tagging attempts using a remotely-deployed suction-cup tag. Mar. Mamm. Sci. 14:316–324.

Smolker, R. A., A. F. Richards, R. C. Connor, and J. W. Pepper.
1992. Sex differences in patterns of association among Indian Ocean bottlenose dolphins. Behaviour 123:38–69.

Walker, B. G., and P. L. Boveng.
1995. Effects of time-depth recorders on maternal foraging and attendance behavior of Antarctic fur seals (*Arctocephalus gazella*). Can. J. Zool. 73:1538–1544.

Weinrich, M. T., R. H. Lambertsen, C. S. Baker, M. R. Schilling, and C .R. Belt.
1991. Behavioural responses of humpback whales (*Megaptera novaeangliae*) in the southern gulf of Maine to biopsy sampling. Rep. Int. Whal. Comm. Spec. Issue 13:91–97.

Weinrich, M. T., R. H. Lambertsen, C. R. Belt, M. R. Schilling, H. J. Iken, and S. E. Syrjala.
1992. Behavioral reactions of humpback whales *Megaptera novaeangliae* to biopsy procedures. Fish. Bull. 90:588–598.

Weller, D. W., V. G. Cockcroft, B. Wursig, S. K. Lynn, and D. Fertl.
1997. Behavioral responses of bottlenose dolphins to remote biopsy sampling and observations of surgical biopsy wound healing. Aquat. Mammals 23:49–58.

White, G .C., and R. A. Garrott.
1990. Analysis of wildlife radio-tracking data. Academic Press, San Diego, CA, 383 p.

Whitehead, H., J. Gordon, E. A. Mathews, and K. R. Richard.
1990. Obtaining skin samples from living sperm whales. Mar. Mamm. Sci. 6:316–326.