

Behavioral responses of humpback whales, *Megaptera novaeangliae* (Cetacea: Balaenopteridae), to satellite transmitter deployment procedures

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ABSTRACT. Tagging whales with implantable satellite transmitters creates the possibility of disturbing the animals. Between 2003 and 2005, behavioral observations of humpback whales during tag deployment operations were conducted off the coast of Brazil from the flying bridge of a speedboat. The speed achieved by each whale during pursuit was registered by GPS receivers onboard two inflatable zodiac boats. Respiratory frequencies were significantly lower ($n = 15$, $p < 0.05$) before (mean = 0.89 ± 0.06 blows/min) compared with after (1.39 ± 0.15) tagging. The same effect was observed for the speed of each animal (mean = 10.96 ± 0.44 km/h and 12.54 ± 0.57 km/h; $n = 13$, $p < 0.05$). Both variables were positively correlated with the duration of the pursuit ($n = 21$, $r = 0.88$, $p < 0.05$; $n = 13$, $r = 0.94$, $p < 0.01$) and with each other ($n = 26$, $r = 0.65$, $p < 0.01$). Acute responses were observed in 50% of the 28 tag deployments. Pursuits were shown to generate a longer effect than tagging. We suggest that the behavioral changes presented here are short-term disturbances because the tagging operation ends quickly and is not a repeated procedure. However, protocols must be developed to guarantee the maintenance of the animals' welfare during operations.

KEY WORDS. Behavior; respiratory frequency; satellite telemetry.

There has been growing concern over the last few decades about the effects of environmental stress, including anthropogenic factors, on marine mammals (FAIR & BECKER 2000). However, only a few publications have reported on the nature or scale of the impact caused by scientific experiments. Hence, it is not possible to quantify the effects of research using the existing literature (GALES *et al.* 2003).

It is important to recognize behavioral changes when monitoring animal behavior because these changes could signal a decline in the welfare of the subject. The use of behavioral measures to evaluate welfare was emphasized by DAWKINS (2003). Behavioral records can be an important early indicator of health problems, and may also serve as a useful tool for recognizing potentially stressful social changes and circumstances (WAPLES & GALES 2002). The study of respiratory frequency is also important in evaluating animal welfare, especially for animals in which physiological measures are difficult (BROOM & JOHNSON 1993), as in the case of cetaceans.

The humpback whale, *Megaptera novaeangliae* (Borowski, 1781), has been the most intensively studied of the mysticete

whales, and it is one of the world's most wide-ranging mammals (CLAPHAM 2000), found in all of the world's oceans. Some studies have used satellite telemetry to gain more knowledge about the species (e.g., MATE *et al.* 1998; ZERBINI *et al.* 2006). This is considered to be an invasive method, so it is essential to adopt a work protocol that respects the animals' welfare and limits the level of disturbance. To create such protocols, we must know the impact of the satellite-tagging procedures on the observed species.

The main objective of this study was to evaluate the observable reactions of humpback whales to satellite-tagging procedures, including close distance pursuit, in order to obtain data that may be relevant in the design of future research protocols.

MATERIAL AND METHODS

The study was conducted in October and November of 2003, 2004 and 2005, between 18 and 35 nautical miles from the coast of the states of Espírito Santo and Bahia, Brazil. A speedboat was used as an observation platform to record most of the behavioral data. Two zodiac boats were used to approach, tag,

and register the behavior of the whales. The zodiacs used 50 or 90 hp engines, and measured 4.5, 5.5 or 6.5 m, but the effects of different propulsion systems and zodiac dimensions were not analyzed in this paper. The transmitter (Wildlife Computers – Redmond, WA, USA) and fixation system (Greenland Institute for Natural Resources) configuration used in most cases (called short implantable) consisted of a stainless steel pole weighing 222 g and measuring 42 cm and 2 cm in diameter. The antenna was 14 cm long, and the total length of the device was 28 cm when the transmitter and anchoring system were included. Approximately 24 cm measured the maximum length that could be inserted into the animal integument (fat layer and probably muscle layer). We also used some longer (47 cm) and heavier (260 g) transmitter configurations, but we did not analyze the effects of different transmitter configurations.

In order to tag an animal, a zodiac boat approached the whale laterally, positioning itself less than 4 m from the animal. Tagging was conducted using carbon and fiber poles that measured 6-8 m, with an inserter on the anterior end, where the transmitter and biopsy sampler were attached. Aerial behaviors (lobtailing, acceleration, fast submersion and speed) and respiratory frequency (time intervals between blows) were registered during the process. When an animal or group was observed, the boats moved towards their location. Whales were then pursued and behavioral data were registered. The zodiac boat responsible for the tagging kept a distance of 3-40 m from the whale group or solitary individual, and the second zodiac boat kept a small distance (generally <50 m) from the first. The speedboat maintained a larger distance, generally a few hundred meters from the pursuits, but was generally within sight of the whales and zodiacs. When the speedboat was out of sight, behavioral recordings (except GPS register – explained in more detail below) were terminated. The duration of the pursuits before tagging varied, and animals were tagged as soon as possible, depending on various factors, such as speed, swim behavior, oceanic conditions and other factors. The time intervals between blows were registered using a stopwatch and the Focal Animal Method (ALTMANN 1974). During the sampling period all of the respiratory expirations (blows) of one animal were registered. This behavior was observed before and after the tagging. After tagging, pursuit continued in order to register the position and situation of the transmitter on the body of the animal, but from a greater distance (>20 m) from the animal(s). These pursuits varied during the study and lasted only for as long as it was required to register the position of the tag.

The speed of each whale during pursuit was registered using a GPS receiver in each zodiac, set to automatic mode. Observable behaviors (lobtailing, acceleration, fast submersion) displayed during tagging were also registered using the Focal Animal Method (ALTMANN 1974) and were labeled as “acute responses to tagging.” Twenty-eight acute tagging responses were analyzed for four escorts (males), one solitary female and 23

animals swimming together with calves (six females – genetic analysis). Owing to the large number of females and the small number of males, analyses focusing on comparisons between sexes and different group compositions were restricted.

Data were analyzed using the Kolmogorov-Smirnov normality test. The values obtained for the factors analyzed here (respiratory rate, speed and observed aerial behaviors; $p < 0.01$) indicated the need to employ non-parametric analyses. The standard error was calculated for each mean. Sample sizes (n) varied among the different data groups, reflecting differing sampling conditions. It was rarely possible to sample respiratory frequency, speed and acute responses in the same time for each sampling situation. The Wilcoxon test was used to evaluate whether there was a significant difference between the mean values obtained for respiratory rate (respiratory frequency) and also the speed of the animal (compared before and after the tagging). For each subject, respiratory rate was calculated by dividing the number of blows by the duration of sampling in minutes.

Respiratory frequency and speed were grouped into 5-minute intervals, and mean values were calculated for each interval. The correlation of respiratory frequency and speed of pursuit with the duration of the pursuit was analyzed using the Spearman test. In the cases of simultaneous registration of respiratory frequency and speed, the data were paired and tested for correlation using the Spearman test. The acute response behaviors (those presented immediately after tagging) and the absence of a visible response were registered. The Mann-Whitney test was used to compare two behaviors and the Kruskal-Wallis test was used to compare three or more behaviors.

RESULTS

Table I shows the mean pursuit duration of the humpback whales obtained during this study and the mean duration of expiratory blows. The maximum duration of a pursuit was 1.35 hours, with two interruptions of approximately five minutes each. The maximum duration of an uninterrupted pursuit was 51.75 minutes.

Mean respiratory rates, measured before ($n = 15$, mean = 0.89 ± 0.06 blows/min) and after tagging (1.39 ± 0.15), differed significantly ($p < 0.05$). This shows that the mean respiratory rate before tagging was lower than the mean rate registered after tagging. Mean speeds before (10.96 ± 0.44 km/h) and after (12.54 ± 0.57 km/h) tagging were also significantly different ($n = 13$, $p < 0.05$). Table II shows the mean values of respiratory frequency and animal speed. It presents a clear representation of the behavioral changes registered after tagging. Whale respiratory frequency was positively correlated with the duration of pursuit ($n = 21$, $r = 0.88$, $p < 0.05$, Fig. 1).

We observed a gradual increase in the mean speed of the whale during the initial 30 minutes of pursuit, increasing from 8 ± 0.57 to 10.83 ± 0.8 km/h. Correlation analysis ($n = 13$, $r = 0.94$, $p < 0.01$) demonstrated that the mean speed was posi-

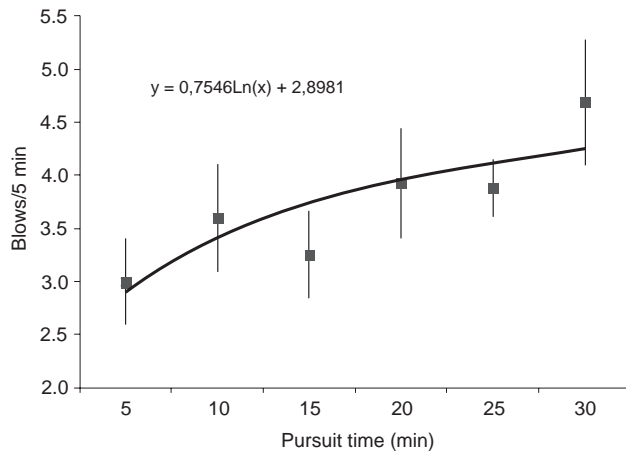


Figure 1. Respiratory frequency of the humpback whales during pursuit and respective tendency line and logarithmic equation, showing a significant gradual increase.

tively correlated with the duration of pursuit. An apparent stabilization in speed occurred after the first 30 minutes ($n = 13$, $p > 0.05$) and continued until the end of the sampling period (55 min). The mean speed after 30 minutes of pursuit was 13.24 ± 0.35 km/h. The maximum speed observed was 16.80 km/h, which was registered after approximately 30 minutes of pursuit. When analyzing the whole period, we found that speed was positively correlated with the duration of the pursuit ($n = 13$, $r = 0.85$, $p < 0.05$, Fig. 2).

Mean speed and respiratory rates were found to be positively correlated when analyzed in 5-minute intervals, up to 30 minutes of pursuit ($n = 26$, $r = 0.65$, $p < 0.05$, Fig. 3). The following behaviors were observed during tagging (acute responses): lobbaiting (0.214 ± 0.078 , or 21.4%), acceleration

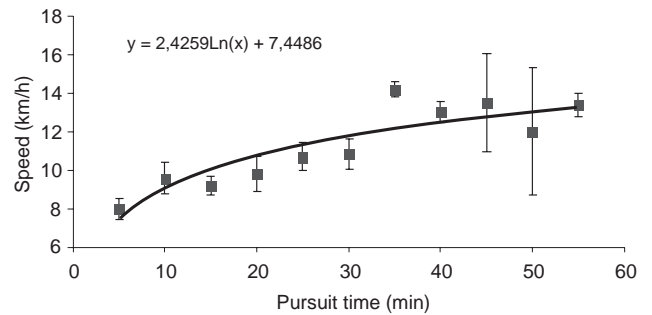


Figure 2. Mean speed and pursuit time, and respective tendency line and logarithmic equation registered in this study. A positive correlation was seen between the two measures in the initial minutes, and the mean speed stabilized after 30 min of pursuit.

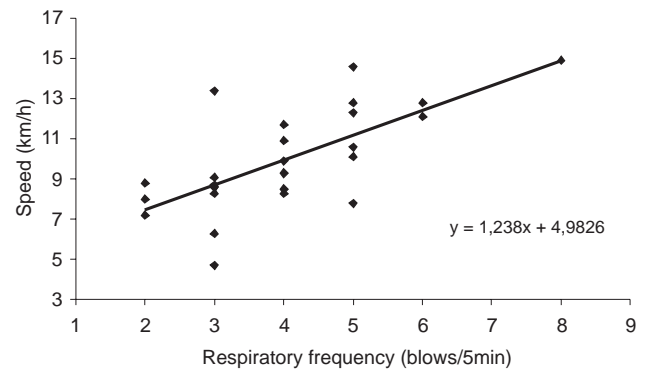


Figure 3. Positive correlation between speed and respiratory frequency of the humpback whales during this study, and respective tendency line and linear equation. The highest value observed for the respiratory frequency was 8 blows/5 min (1.6 blow/min), with a mean speed of 14.9 km/h.

Table I. Mean duration of pursuit and respiratory behavior (r.b.). Total and partial duration mean, before and after tagging.

	Mean duration of pursuits (min)	Min – Max (min)	R. b. samplings mean (min)	Min – Max (min)
Before tagging	22.48	4.00 – 69.78	18.86	3.93 – 57.98
After tagging	9.00*	1.00 – 27.87	9.23**	2.81 – 26.37
Total	27.18	4.78 – 81.10***	24.01	4.78 – 70.83***

*All pursuits included. **Only pursuits where respiratory behavior sampling occurred. *** In some occasions, there were pursuit or r.b. data collected only before or after tagging.

Table II. Mean values of respiratory frequency and speed, before and after tagging, and the respective sample sizes (n) of different group compositions analyzed. Speed was measured for the whole group, so the escort category was excluded.

	Before	After	n	Mother and calf*	Solitary	Escort	Mother, calf and escort*
Resp. freq. (blows/min)	0.89	1.39	15	12	1	2	0
Speed (km/h)	10.96	12.54	13	7	0	–	6

*Probable mothers.

(0.214 ± 0.078 , or 21.4%), fast submersion and acceleration (0.035 ± 0.035 , or 3.5%), and lobtailing and acceleration (0.035 ± 0.035 , or 3.5%; Fig. 4).

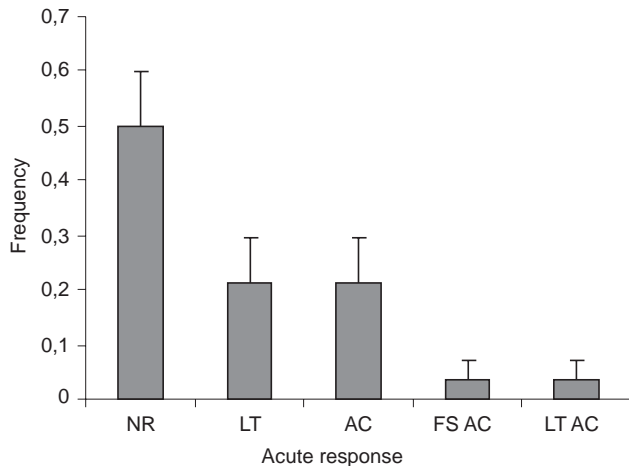


Figure 4. Frequency of no responses (NR) and observed responses (LT = lobtailing, AC = acceleration, FS AC = fast submersion followed by acceleration, and LT AC = lobtailing followed by acceleration) executed by the humpback whales immediately after tagging (acute responses).

The mean incidence of acute behaviors was variable ($n = 28$, $p < 0.05$). In 50% of the cases, there was no immediate reaction to tagging (NR, mean = 0.5 ± 0.096). In two cases, we observed two consecutive behaviors: fast submersion and acceleration, and lobtailing and acceleration. Tail breaching was observed once, but has been included in the lobtailing category. Two of the four male escorts responded to tagging, as did the solitary female.

DISCUSSION

The present study has found a significant change in the respiratory frequency of tagged whales in the periods immediately before and after tagging. Additionally, the gradual change in this parameter and speed during the initial minutes of pursuit suggests that the act of pursuing the animals influenced the results. Thus, tagging itself was not the only factor disturbing the animals. We suggest that the animals maintain high respiratory frequencies and speed during pursuit. As with heart rate, an increase in respiratory frequency can be a response to a situation perceived by the animal as stressful (BROOM & JOHNSON 1993). MOBERG (2000) emphasizes change in respiratory frequency as one of the probable responses to disturbances.

Humpback whale respiratory behavior and speed were considered by us to be sufficient to evaluate pursuit effects. The

same parameters had been previously evaluated for fin whales, *Balaenoptera physalus* (Linnaeus, 1758), in the Mediterranean Sea, *Balaenoptera physalus* (Linnaeus, 1758), by JAHODA *et al.* (2003). The authors used respiratory frequency and speed to evaluate the additional impact of the approach of a small speedboat on a whale that was already being followed at a distance by a larger vessel. Significant alterations in both parameters were observed, indicating that the animal was feeling disturbed.

A possible explanation for the stabilization of the animal's speed after 30 min of pursuit is that this is the mean time it takes for the humpback whale to reach physical exhaustion. Despite continued pursuit after tagging, this stabilization could also be explained by the fact that, after tagging was completed, the vessels maintained a larger distance from the whale(s). Because the main objective of our study was to deploy the satellite transmitters, we did not evaluate which factor (whale exhaustion or distance from pursuing vessel) accounted for speed stabilization.

The high speeds registered for the whales in this study suggest that the increase in speed was a direct response to the pursuit itself. These results are consistent with SCHEIDAT *et al.* (2004), who observed that humpback whales reacted to the approach of whale-watching boats by increasing their speed significantly (from 2.97 km/h to 4.52 km/h during the vessel interaction). Furthermore, in a study conducted by FÉLIX (2004), the average speed estimated for 76 humpback whale groups moving off the coast of Ecuador was 4.53 km/h, substantially lower than the speeds observed in the present study. According to BROOM & JOHNSON (1993), escape and withdrawal are indicators of disturbance.

In half of the events we analyzed, the animals did not respond to the tagging. Despite the fact that the transmitters used were larger in comparison with biopsy darts used in the studies cited below, the animals were already displaying escape behavior when they were tagged, as shown by their increased speeds, perhaps inhibiting aerial behavioral responses that could cause a decrease in speed. It is also possible that lobtailing in response to tagging is a reflex since the frequency and type of reaction was small and independent from the dimensions of the equipment.

BROWN *et al.* (1994) observed detectable reactions (e.g.: lobtailing) to biopsy sampling using a crossbow and floating arrows in 41.6% of the humpback whales they observed in Australia. In a similar study conducted in Silver Bank (Dominican Republic), CLAPHAM & MATTLA (1993) observed small reactions in 22.5% of the animals they observed, and medium to strong reactions in 33.4% of them. Both studies concluded that biopsy sampling produces minimum effects on *M. novaeangliae*. In another study, GAUTHIER & SEARS (1999) demonstrated that the humpback's reaction to biopsy sampling was generally a strong lobtail, and the animals returned to their previous behavior either immediately or a few minutes after the procedure. The acute responses observed in this study suggest that,

in addition to the pursuit, tagging itself is also a source of disturbance, but satellite transmitter tagging happens quickly and is not repeated. According to BROOM & JOHNSON (1993), the impact of a disturbing stimulus on an animal is critically affected by how long the stimulus lasts and how often it occurs.

WEINRICH *et al.* (1992) used respiratory and behavioral measures to evaluate disturbances caused by biopsy sampling in humpback whales. They concluded that the animals' responses to the procedure did not, for most part, differ from their natural behaviors, but that the frequency of certain behaviors changed. They proposed that the only exception was the hard tail flick (lobtailing), which was rarely observed in any situation other than a response to the biopsy sampling. In the present study, lobtailing was observed in response to the tagging and also during pursuit. In the Weinrich study, whales were approached by a relatively silent sailboat half of the time, and when the engine was turned on, speeds were slow and vessel movement was limited. Tagging whales with poles requires getting close to the animals generally after fast pursuits, causing auditory and visual disturbances. Avoiding fast pursuits would greatly increase the duration of field work, consequently increasing the total cost of the research, but would certainly cause a significant decrease in the disturbance caused by tagging. Animal groups that do not exhibit calm behavior (e.g., resting, like most of the groups observed during this study; animals presenting calm behavior are easier to tag) would be ignored by the taggers and would not be pursued. The use of a protocol that respects the welfare of the subjects, coupled with continuous behavioral monitoring during tagging activities, can help prevent significant disturbances. Tagging with air guns, which do not require a close approach and fast pursuit, could be an option, but involves more risks and increased monitoring of the operations.

WEINRICH *et al.* (1992) stated that their results were generally consistent with other studies on the reactions of baleen whales to a variety of human-induced stimuli. Like some of the authors cited in this paper, we have found that animals avoid sources of disturbance. Furthermore, we have demonstrated that boat pursuits and tagging increase the animal's level of disturbance. However, the relative contribution of the different factors analyzed is difficult to measure.

In this study we observed several behaviors in response to tagging: acceleration, lobtailing, fast submersion, a combination of fast submersion and acceleration, and a combination of tail slapping and acceleration. BAIRD *et al.* (2004) analyzed the behavior of two Ziphiidae individuals to tagging using remotely-deployed suction-cup attached transmitters. They observed fast submersion – *Ziphius cavirostris* Cuvier, 1823, and a combination of tail slapping and fast submersion – *Mesoplodon densirostris* (Blainville, 1817), a similar pattern to that exhibited by humpbacks. The authors suggested that reactions to tagging are only short-term.

We did not observe any attempts to remove the implanted

transmitter, or differences in the behavior of the tagged whales when compared with other members of their groups. Because the transmitters were inserted deep into whale's integument, they most likely did not cause any significant hydrodynamic drag or disturbance in social interactions.

Our study showed that satellite tagging procedures caused increases in the respiratory frequency and speed during pursuit. The combination of pursuit and tagging generates a net disturbance effect, causing the animal flee from the sources of stimuli. Pursuit probably generated a longer effect duration compared with tagging. We suggest that the behavioral changes described here are a response to short-term disturbances, because they end quickly and are not repeated. Nevertheless, protocols that guarantee the maintenance of the welfare of the subjects during experiments should be followed. Here we suggest that the pursuit time during satellite tagging procedures be restricted to 30 min.

Despite logistic restrictions, it is important, in the future, to register parameters like aerial and respiratory behaviors in situations before and after disturbances, to evaluate changes in behavior due to the procedures and the return to the original behavior.

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