Mitigation of harm during a novel behavioural response study involving active sonar and wild cetaceans

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ABSTRACT

Some studies of how human activities can affect wild free-ranging animals may be considered to have potential negative outcomes too severe to be ethically studied. This creates a societal dilemma involving choices between continuing risky activities with high uncertainty about their potential effects on wildlife, often with considerable associated precaution or undertaking focused research to reduce uncertainty, but with some risk of harm from either strong response leading to potential stranding or direct physical injury from sound exposure. Recent and ongoing field experiments have measured the conditions in which wild cetaceans respond to military sonar, and provided insight into the nature of responses. Here mitigation measures are reported for one of the first such experiments designed to measure fine-scale behavioural responses to controlled exposures of mid-frequency (3–4 kHz) active sonar. The objective was to do so without causing the kinds of physical harm that have been previously observed (e.g. stranding events) and that motivated the study. A critical goal of this experimental study was to identify a response that was safe but that could be used as an indicator of the probability of risk from more extreme or sustained exposure from real military operations. A monitoring and mitigation protocol was developed using a feedback control procedure for real-time mitigation of potential harm. Experimental protocols were modulated relative to indicators of potential risk with the explicit objective of detecting potentially harmful consequences of sound exposure and taking appropriate corrective action. Three categories of mitigation methods were developed and integrated within the experimental protocol incorporating designed, engineered, and operational mitigation measures. Controlled exposure experiments involving free-ranging animals were conducted without any evident harm to the experimental subjects, while successfully eliciting behavioural responses that provided meaningful results to inform management decisions. This approach demonstrates the importance of careful design of protocols in exposure-response experiments, particularly in pioneering studies assessing response where both the potential for harm and level of uncertainty may be high.

KEYWORDS: ACOUSTICS; CONSERVATION; BEHAVIOUR; MANAGEMENT PROCEDURE; SHORT-TERM CHANGE; BEAKED WHALES; DELPHINIDS; NORTHERN HEMISPHERE

INTRODUCTION

Scientific research plays a key role in understanding the effects of human activities on wildlife and ecosystems. An ethical approach to the management of protected species requires those who undertake the experimental studies involving potential or actual harm to animals to implement best practices in assessing potential trade-offs associated with their work (Farnsworth and Rosovsky, 1993; Gales *et al*., 2010). Careful and deliberate measures must be taken to reduce the number of animals that will be disturbed and to minimise the amount of pain and suffering required to obtain scientific results. If specific research procedures pose a risk of harm to individuals, it may only be justifiable when there are sufficient, identifiable benefits for effective conservation and management (Boyd, 2002). This study looks at ethical issues of research designed to protect wild animals from poorly understood human risks. They were assessed in a situation where it was difficult to guarantee protection of subjects in the wild. It was also impossible to accurately estimate (in advance) the number of whales required to guide management decisions.

The particular case discussed here involves several species of beaked whales (*Mesoplodon* sp. or *Ziphius cavirostris*) for which lethal strandings have been reported to coincide with naval sonar exercises (D'Amico *et al*., 2009). Cox *et al*. (2006) reviewed potential mechanisms by which sonar might harm these whales. They conclude that physical effects of sound on tissue, which could be studied in tissue *in vitro*, requires sound levels so high that it is unlikely to initiate strandings. They conclude that anthropogenic noise may in some conditions elicit a behavioural reaction that may disrupt diving physiology and lead to strandings. These behavioural reactions can realistically only be studied with beaked whales at sea. The challenging goal for this study was to identify a response to sonar that was safe for the subject, but could also indicate risk of stranding if exposure were longer and/or more intense, and to quantify the exposure conditions required to elicit the response.

This study is the first to directly examine the behavioural mechanisms underlying these adverse effects of the specific types of mid-frequency active (MFA) military sonars involved in previous stranding events with cetaceans, especially beaked whales. The experiment was conducted on a Navy training range in the Bahamas and involved a controlled exposure experiment (CEE) paradigm (see Tyack *et al*., 2011 for full details of the playback stimuli). This experimental approach can test which sound exposures actually cause behavioural effects, a test that may not be possible in observational research. Opportunistic observations during actual (uncontrolled, non-experimental)

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sonar events provide some general insight into understanding of behavioural responses. However, CEEs occur within an experimental paradigm to allow the collection of adequate pre-exposure behaviour measurements, the ability to carefully control sound source output characteristics and location relative to experimental subjects in order to achieve a desired range of sound exposures. For the collection of post-exposure behaviour in particular, a dose-escalation protocol was used, which can identify the lowest sound exposure level that elicits a particular behavioural response.

Before this Behavioural Response Study (BRS) took place, the type and magnitude of potential responses of individual whales to exposure of simulated sonar, especially MFA, and other sounds were largely unknown. Thus, a highly precautionary approach was required to evaluate and mitigate harmful impacts from the experiment by using an adaptive design to enable rapid response to negative indicators. This study agrees with Farnsworth and Rosovsky (1993) that the scientific community should incorporate more explicit discussion and evaluation of ethical issues associated with field ecology experiments, but this study also required evaluation of ethical issues by outside bodies as well. This study involved marine mammals, so the planning for the study required evaluation of these issues in applications for approval by bodies external to the study team, as required by combinations of the funding organisations, federal or local regulatory requirements, and the requirements of participating organisations. These included the Office of Protected Resources of the US National Marine Fisheries Service, which issues permits for scientific research on marine mammals, a US Institutional Animal Care and Use Committee (Woods Hole Oceanographic Institution), and a UK Animal Welfare and Ethics Committee.

The present analysis considers the effectiveness of an operational control procedure employed in Tyack *et al.* (2011) involving the playback of three different sound stimuli: (i) a simulated mid-frequency naval sonar signal (MFA) with both constant frequency and frequency modulated tonal components in the 3–4kHz band; (ii) a pseudo-random noise signal (PRN) with overall bandwidth and timing similar to simulated MFA; and (iii) killer whale (ORCA) sounds from wild marine mammal eating (transient) killer whales (*Orcinus orca*). Blainville's beaked whales (*Mesoplodon densirostris*) and several species of small cetaceans (short-finned pilot whales, *Globicephala macrorhynchus*; false killer whales, *Pseudorca crassidens*; and melon-headed whales, *Peponocephala electra*) were the subjects of these experimental exposures. The beaked whales were selected as the primary species identified as sensitive to sonar. Delphinids were included as a series of comparison species with differing social structures to test their relative sensitivity to the beaked whales, and whether differential social responses to potential threats might affect the probability of flight reactions and potential associated risk of stranding.

Given the objectives for studying these aspects of behaviour in an experimental context, but recognising the potential for responses that could result in harm to these species, some of which had been involved in previous stranding events involving actual MFA sources, the integrated and adaptive mitigation strategy described here was designed. This strategy included specific integrated and adaptive elements both in the planning, implementation, and evaluation of noise exposure and response. Mitigation measures were included in the overall experimental design (e.g. site selection, testing conditions), engineering of experimental protocols (e.g. source ramp-up), and operational implementation of mitigation in different experimental modes (e.g. source shut-down, post-hoc visual surveys of the study area). Particularly the operational measures are integrated to provide multi-variable data (e.g. visual surveys, real-time passive acoustics) on the distribution and behaviour of experimental and other subjects in order to effectively monitor the experiment to ensure successful testing of responses while mitigating any potential harm. While they may have some broader implications, the resulting protocols and data from this study are particularly relevant to the informed and adaptive development of experimental design and potential real-time mitigation for studies of the effects of real sonar operations on cetaceans.

METHODS

The Tyack *et al.* (2011) study took place in July–September 2007 and 2008 in the Bahamas. The study was conducted under marine mammal research permits issued by the US National Marine Fisheries Service to John Boreman (Permit No.1121-1900; B. Southall was the designated principal investigator) and to Peter Tyack (Permit No.981-1578), and issued by the Government of the Bahamas to the Bahamas Marine Mammal Research Organization (Bahamas permit No.01/09) and Ian Boyd (Bahamas permit No.02/07 and No.02/08). The study was carried out in strict accordance with the conditions of these permits and the US Animal Welfare Act following the recommendations of the Guide for the Care and Use of Laboratory Animals of the National Institutes of Health (Clark *et al*., 1996); protocols were also approved by the Institutional Animal Care and Use Committees of the main participating institutions.

A strategically integrated, multi-faceted monitoring and mitigation protocol was developed with the explicit objective of detecting potentially harmful consequences of experimental trials and taking appropriate corrective action in an informed and adaptive manner before, during, and following experiments. To meet this objective, three categories of mitigation methods were developed and integrated within the experimental protocol, incorporating designed, engineered and operational mitigation measures. Research took place within an operational control procedure with clearly specified lines of communication and responsibility (Fig. 1).

Designed (pre-experimental) mitigation

Site selection

The field site was to the east of Andros Island, Bahamas in the Tongue of the Ocean (24.0903°N, 77.2350°W), a deep water basin surrounded by islands and sand banks. This site was selected because of the presence of the study species and the demonstrated capability to detect and locate beaked whales acoustically using the US Navy's Atlantic Undersea Test and Evaluation Centre (AUTEC), an underwater acoustic range (DiMarzio *et al*., 2008). AUTEC had 82

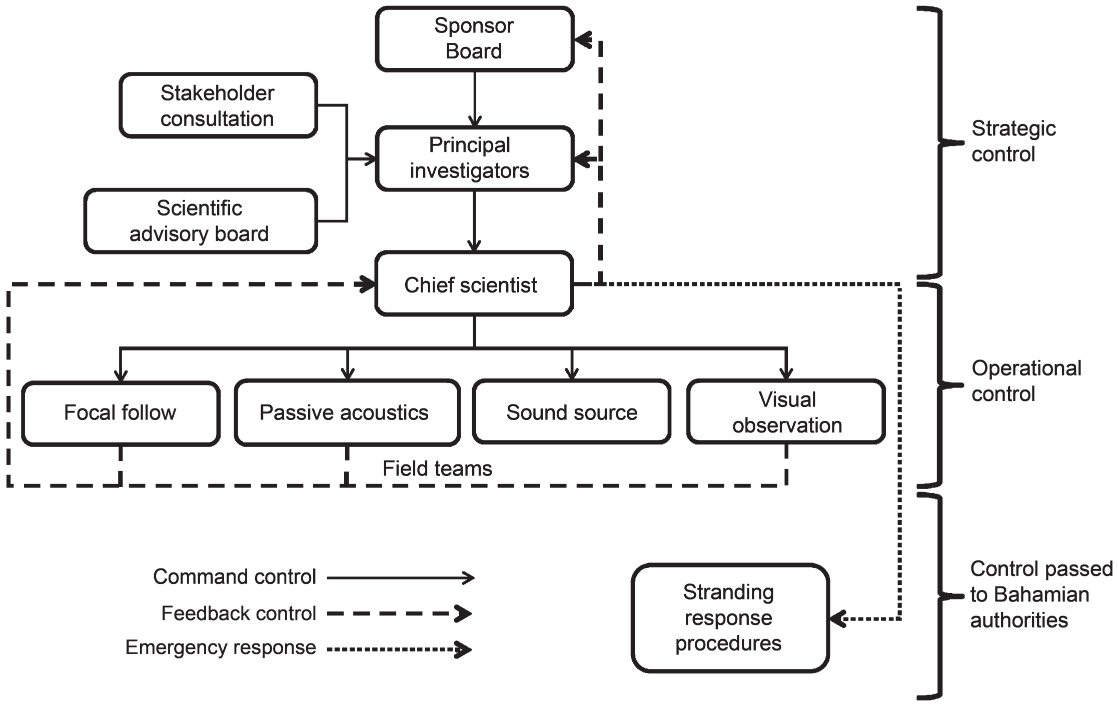

Fig. 1. Flow of communication and control in the strategic and operational management of the experimental design. This process involves the command control process as well as the flow of information that provides feedback control to the management team from those gathering information in real-time. Not shown here are the real-time data audit procedures that were designed to allow a *post-hoc* detailed analysis of the activities that could have resulted in the death or stranding of animals.

hydrophones mounted in a grid at roughly 4km spacing on the seafloor at depths of ≤2,000m that were cabled back to shore. All playbacks took place within the boundaries of the underwater range to allow continual real time acoustic tracking and mitigation, as has been demonstrated during military training exercises (McCarthy *et al*., 2011).

Observation time and space scales

Three time- and space-scales were used for obtaining observations that allowed the assessment of the effects of experiments (Table 1).

Selection of environmental conditions

Since locating, observing and tagging focal animals was not possible in high sea states, sound playbacks were not conducted if Beaufort Sea state was > 3. Similarly, hours of darkness (or periods of low visibility), were avoided as much as possible by not conducting playbacks after midday.

Engineered (experimental sound source) mitigation

Engineered measures primarily focussed on the sound source output. The signal used by Tyack *et al*. (2011) was a simulation of a typical operational US Navy mid-frequency sonar signal, which had an initial 0.5s linear frequency-modulated upsweep from 3.5–3.6kHz, followed by a 0.5s constant frequency tone at 3.75kHz, a 0.1s silent period, and then a 0.5s constant frequency tone at 4.05kHz. The total duration was thus 1.6s, with a repetition rate of every 25s from the onset of one signal to the onset of the next. The custom source used had a maximum source level of 211–212dB re 1µPa@1m. The distance between the source and focal animal was adjusted to ensure the received level did not exceed 160dB re 1µPa@1m. This was a level that was not expected to cause any temporary or permanent hearing threshold shifts based on a very conservative interpretation of the available data on auditory impacts of noise exposure available at that time (Southall *et al.,* 2007). The acoustic engineers who utilised the AUTEC range for marine mammal monitoring had experience with the propagation of mid-frequency sonar signals in the study area. They used standard parabolic equation and Bellhop sound propagation models with the sound source output and known features of the AUTEC range (e.g. bottom type) to model the range of predicted received levels for multiple depths in the water

Table 1

Representation of the three time and space scales covered by different platforms using specific modes of observation that provided feedback of information used in mitigation.

Scale	Space	Time	Mode	Platform
Large	10–> 100km	Days–weeks	1. Aerial survey 2. Acoustic array	1. Twin engine aircraft 2. AUTEC hydrophones
Medium	0–10km	Hours–< 1 day	1. Ship-based elevated platform using big-eye binoculars 2. Acoustic array	1. > 30m vessel 2. AUTEC hydrophones
Small	10m–1km	Seconds–hours	Focal follow of exposed animals	< 6m vessel

column around possible positions of the sound source. It was assumed that animals could be at any reasonable depth based on their species-typical dive behaviour that corresponded with the highest received levels. This information was used to determine appropriate source-animal ranges to meet the experimental goals without exceeding this target received level. Propagation modelling was conducted *a priori* for possible areas where experiments could occur to identify any potential site-specific differences. In addition, a ramp up of the amplitude of the sound was performed as part of the dose-escalation protocol, and as required under conditions of the research permit (see Tyack *et al.,* 2011 for details).

Operational (experimental and post-experimental) mitigation

The actual conduct and post-exposure monitoring of the experiment took place under five integrated and adaptive operational modes, each defining different sets of activities. The explicit distinction of these modes and the mitigation measures being employed ensured clear communication and a coordinated approach between the Chief Scientist and all teams (Fig. 1). The overall adaptive approach involved the use and integration of information from all of the available information from the various field teams (e.g. tagging, visual survey, passive acoustic) by the Chief Scientist in order to ensure the experiment met the mitigation goals and requirements. While elements of the operational mitigation used here were not novel (e.g. source shut-down), the integration of various data streams and the dynamic nature of visual data for surface animals transitioning to acoustic data in real time for diving animals with a seamless, real-time transition between these tools based on the behaviour of experimental subjects was a unique development in this study. The operational procedure progressed sequentially from one mode to the next, with each mode having associated and adaptive operational mitigation measures:

(1) *Search and assessment* involved the localisation of candidate whales for a CEE. Initial identification was carried out either acoustically using the AUTEC hydrophone sensors or by visual observers with subsequent photo-identification of individual animals. Mitigation measures aimed to identify all animals prior to the CEE, to ensure that no single animal was exposed more than once, and that a suitable age class animal was chosen (Table 2);

(2) *Tagging* involved attachment of acoustic and movement loggers (DTAG – see Johnson and Tyack 2003, Tyack *et al.*, 2011) on focal animals. Mitigation measures ensured that tagging was completed by experienced personnel to minimise approach attempts and ensure good tag attachment (Table 3);

(3) *Playback* involved sound source operation. Mitigation measures were implemented to ensure that maximum source levels were not exceeded and ramp up protocols were followed (Table 2). Shut-down mitigation (all sound transmission immediately ceased) was applied if any animals were observed within 200m of the source vessel (a required permit condition to prevent any potential physical injury or animals coming very close to the sound source). This was determined through range finding binoculars or estimated by the naked eye. Source

Table 2

Actions in the sequence carried out during playback experiments in 2007.

Stage	Action
Start:	
Pre-start preparation	Cast XBT if required to measure sound speed profile Transmit XBT data to modellers Run sound propagation loss model Decide on source depth based upon thermocline depth
Focal follow group of whales	Determine whether a juvenile is in the group Photo-identification of each whale in the group to determine whether any have been exposed before
Initiate post-exposure monitoring and mitigation procedure	Alert aircraft for deployment for aerial survey
Spatial disposition of platforms	Ensure vessel carrying the sound source is 1,000–2,000m from whales Ensure no other whales are within 200m of the vessel using 360° sweep with bigeye binoculars
Playback shutdown:	
Passive Acoustic Monitoring (PAM)	Cessation of clicking in beaked whales determined by the AUTEC array Unusually rapid movement or vocalisations
Visual observers	Strong and abnormal directed swimming (at surface) Increased and abnormal surfacing rate and respiration rate Animal surfacing with pattern(s) of directed movement, especially toward shore Unusual and abnormal surface/subsurface behaviour involving apparent disorientation and confusion or loss of group cohesion Animal defaecation on an unusual scale during or immediately after playback transmission Focal follow cannot be maintain because of weather Whale sited within 200m zone around the source
Post-exposure monitoring:	
	Track the tagged whale and its associated group with the assistance of M3R until nightfall Survey a region covering a 5km radius around the exposure site Conduct a regional aerial survey including the coastlines

Table 3

Summary of monitoring and mitigation effort during BRS divided down by each team and activity.

Team	Personnel	Location	Main activity	Region of operation	Monitoring and mitigation responsibility	Communication
Visual observers	Two teams of four, on a 30 minute rotation, each with an experienced team leader, 2 individuals scanning with 25 × 150 big-eye binoculars, 1 with 7 × 50 binoculars and 1 was in charge of communications	On main support vessel that also contained the sound source	Animal detection, tracking and behavioural observation pre-tagging and during and post the CEE. Observations ceased when conditions prevented effective monitoring, at which time radio direction finding on the VHF signals of the Dtag was used to track individuals.	10km radius of ship using 25 × 150 big-eye binoculars	Report unusual behaviour or dangerous occurrences	Team leader in constant communication with own team members via a duplexed 2-way communication system, the Chief scientist and focal follow team.
Focal follow	A team of five individuals under the direction of an experienced skipper	On a small boat	Close up animal detection, tracking, photo-identification and behavioural observation. Follows occurred before, during and after the CEE, until conditions required return to base at which time, radio direction finding on the tag was used to track individuals.	Up to 15km radius of main ship often guided by the Visual observation and the M3R teams	Report unusual behaviour or dangerous occurrences	The skipper was in constant communication with the Chief Scientist and was directed on to the whales by the team leader of the visual observers.
Marine mammal acoustic monitoring on the AUTEC array	A team of experienced acoustic personnel	At the AUTEC base	Animal detection, tracking and acoustic behaviour observation in real-time, before, during and after the CEEs, using detection and localisation (Moretti *et al.*, 2002; DiMarzio *et al.*, 2008; Tyack *et al.*, 2011).	Across the whole AUTEC array using the acoustic monitoring capability of the range	Report changes in behaviour in terms of vocalisations and/or animal movement	Directly with the Chief Scientist during the CEE.
Tagging	A team of one tagger and one boat driver	On a small boat	Deployment of tags, quick look analysis on retrieved tags, in case of requirement to adapt protocol to reduce the potential for harmful impacts on individuals.	Within a few 10s of metres of the focal group	Report unusual behaviour or dangerous occurrences	Directly with the focal follow skipper and Chief Scientist.
Aerial survey	A team of three observers and a pilot	Flights took off and landed at AUTEC	Systematic survey of coastline and appropriate sea areas, along the central section of TOTO, primarily down-current and down-wind of the playback location. Surveys were carried out if abnormal behaviour was observed or if normal baseline behaviour was not observed post-playback, A final aerial survey was conducted on when all CEE trials were complete.	Up to 100km around region of operation	Report dead or moribund animals either floating or stranded	Observations were reported to the Chief Scientist and communicated with all team leaders.

transmissions were sustained if cessation of clicking observed on the AUTEC hydrophones and held through ascent and first shallow dive. These adaptive measures included both visual and acoustic monitoring that allowed information on the location and behaviour of subjects in real time based on where they were in the water column and their behaviour and potential response. These were used to different degrees based on the situation at any one time, with information from both teams provided to the Chief Scientist;

(4) *Post playback* involved visual and acoustic monitoring of the playback area and surveys for stranded marine mammals. Mitigation measures ensured all protocols were rigorously followed (Table 3); and

(5) *Stand-down* was when no CEEs were conducted. Mitigation measures ensured all post CEE monitoring had taken place in accordance with the strategic and operational management design (Fig. 1)

Communication was via an open VHF radio circuit to ensure all teams at all times were in contact with the Chief Scientist. The actions undertaken to ensure mitigation of negative effects of the CEEs are listed in Table 2. Visual and acoustic monitoring by trained personnel (on the sound source vessel, small tagging and focal follow RHIBs, and ashore listening to the acoustic sensors) under the direct communication from an experienced team leader was implemented during all operational modes except 'stand-down'. Operational safety to people and animals was the primary consideration. Protocols to terminate sound transmissions were enacted (Table 2) if animals occurred within a specified physical range of the active sound source or if a potentially harmful response was observed. The NMFS permit (No.1121-1900) governing this research specified that, 'a playback episode must be discontinued if an animal exhibits a strong adverse reaction to the playback activity or the vessel (e.g. breaching, tail lobbing, underwater exhalation, or disassociation from the group).' This requirement was integrated into the mitigation protocols, but additional measures capturing other means of detecting and responding to potential responses were added above and beyond these requirements (see Table 2).

While it was highly unlikely that the limited duration and much lower power (than actual MFA sonar) sound sources, during the first year (2007) of this BRS would cause any direct injury or result in strandings, seven aerial surveys were flown in different contexts to survey for any potentially stranded animals. Two of the flights took place prior to any CEEs, to ensure no existing stranded or dead animals were in the vicinity of the field site. Four were mitigation flights to search the shoreline along the eastern coast of Andros Island, the small islands in North and Middle Bight, the bank edge and cays along the east side of Tongue of the Ocean (TOTO), and the coastal areas of New Providence Island. The final flight was the post BRS monitoring flight. Flights were flown at an altitude of 500ft and at 90 knots. A total of 16.4 hours of flight time was cumulated covering 1,476 n.miles (2,731km). There were three marine mammal sightings; two of bottlenose dolphins (*Tursiops truncatus*) and one of rough-toothed dolphins (*Steno bredanensis*). No sightings of beaked whales were recorded during any flight, based on which what was determined from this form of monitoring to survey for stranded marine mammals was not necessary for the 2008 study. However, as an adaptive approach, when a single flight was called for in 2008 in order to locate the VHF signal of a tag that had detached from a research subject, additional survey effort was added to search nearby beaches for any stranded animals (none were detected – see Table 4).

All operational measures were designed to be adaptive; any information received through feedback control from monitoring and mitigation activities was used to inform decisions (see Fig. 1) on when or whether it was appropriate to proceed in the sequence. In the event of any negative reactions being observed, assessment against the permitted level of disturbance and/or injury was made and reported against the conditions of the issued permits, both in terms of the authorised number and nature of sound exposures relative to predicted sound exposures and in terms of compliance with the required shut-down conditions.

The operational components of the experiment involved teams of researchers under the direction of a Chief Scientist (Fig. 1). The Chief Scientist role was filled by several different individuals at different intervals, but effective communication ensured consistency in decision-making. In 2007, the Chief Scientist was stationed on-shore at a console showing the acoustic data from across the AUTEC range, including the disposition of different vessels involved in the study; in 2008 he was located on the visual observation research vessel (R/V *Roger Revelle*). To achieve the objectives of the monitoring and mitigation strategy, teams were required to work together in a coordinated manner, with each team leading specific activities of key importance to the study (Table 3). Daily team leader meetings were led by the Chief Scientist to discuss any operational changes and the plan for the day.

RESULTS

Seven CEE sequences were conducted during the study. Two of these, one in each of 2007 and 2008, involved beaked whales. Of the remainder, one in 2007 and four in 2008, involved delphinids. In 2007, a playback involving simulated mid-frequency sonar and social calls of transient killer whales was conducted on a tagged female Blainville's beaked whale. In 2008, a group of three Blainville's beaked whales, one of which was tagged, were involved in a CEE, with exposure stimuli of simulated MFA sonar and PRN (for further details see Tyack *et al*., 2011). In 2007, a CEE was also conducted on a group of short-finned pilot whales containing two tagged individuals. During this CEE, playback transmission was temporarily stopped because a group of short-finned pilot whales, not containing the tagged animals entered the 200m shut-down zone around the sound source. In 2008, the first CEE was conducted with a group of 15–20 short-finned pilot whales containing one tagged individual, but the tag was not recovered. The second and third CEEs in 2008 were on two groups of 12 false killer whales, each containing one tagged individual. The fourth was on a group of 12 short-finned pilot whales and approximately 100 melon-headed whales, during which one

Table 4

Details of the operational mitigation measures carried out during the sound exposures in 2007 and 2008, species include short finned pilot whales (Gm), Blainville's beaked whales (Md), false killer whales (Pc), and melon headed whales (Pe).

Date	Species	Exposure shutdown	Post exposure focal follow	Post exposure Dtag data review	Post exposure mitigation flight (time)
2007					
17 Aug.	Gm	Initial exposure terminated due to animals within 200m of source	Immediately post exposure, the 2 whale groups had joined and were moving towards the source vessel; they were subsequently tracked for 12 hours.	No overt or potentially injurious behaviours were observed on the dive records.	18 Aug (03:55 hrs): 0 animals sighted, 0 stranded animals reported.
17 Aug.	Gm		As above.	No overt or potentially injurious behaviours were observed on the dive records.	As above.
02 Sep.	Md		Visual observers sighted the whale five hours after exposure. The ship tracked the tag by tag radio transmissions until it released 14 hours after exposure.	Maximum received level was 150dB re 1µPa. No potentially injurious behaviours were observed on the dive records.	04 Sep. (02:10 hrs): 2 *Tursiops truncatus* sighted, 0 stranded animals reported; 06 Sep. (05:01 hrs): 3 *Tursiops truncatus*, 0 stranded animals reported; 27 Sep. (02:45 hrs): Undetermined number of *Tursiops truncatus*, 0 stranded animals sighted.
2008					
22 Sep.	Gm		One hour. The two groups of whales joined and continued on a southeast direction into the southern part of the range. However, tag failure and poor weather resulted in loss of contact with the individual.	No Dtag data were analysed as the tag was not retrieved. However, the tagged animal was sighted (verified through photo-ID) on the 26 Sep.	Based on 2007 mitigation results, 2008 risk assessments were adapted to reduce the need for mitigation flights after every exposure. However, due to loss of contact with the individual in this case, flights were carried out as a precautionary measure: 0 animals sighted, 0 stranded animals reported.
26 Sep.	Pc		One hour. Whales slowed their speed but remained grouped together. The whales were then observed swimming at speed away from the source vessel.	Maximum received level was less than 160dB re1µPa (RSM). No overt or potentially injurious behaviours were observed on the dive records.	As above; no mitigation flights carried out.
27 Sep.	Md		One hour. Whales continued to show typical behaviour at the surface.	Maximum received level was less than 150dB re1µPa (RSM). No overt or potentially injurious behaviours were observed on the dive records.	As above; no mitigation flights carried out.
28 Sep.	Pc		One hour. Whales continued on their original course at the same speed.	Maximum received level was less than 160dB re1µPa (RSM). No overt or potentially injurious behaviours were observed on the dive records.	As above; no mitigation flights carried out.
29 Sep.	Gm/Pe		Over one hour. Whales continued to travel slowly and exhibited social behaviour at the surface such as lob-tailing and spy-hopping.	Maximum received level was less than 150dB re1µPa (RSM). No overt or potentially injurious behaviours were observed on the dive records.	As above; no mitigation flights carried out.

pilot whale and one melon-headed whale were tagged at the time of exposure. As in the beaked whale CEEs, exposure stimuli consisted of simulated military sonar, killer whale calls and band-limited noise (for further details see Tyack *et al*., 2011).

From all the information available to us, the mitigation measures implemented in the adaptive approach described above were effective. During relatively good environmental conditions, visual observers from elevated platforms on the sound source vessel were able to monitor marine mammals at the surface in the nearby vicinity of the sound source. In one instance, observers implemented a CEE shut-down as specified in the operational protocols when any marine mammal came inside a 200m radius of the active sound source. Visual observers from small boats were also able to monitor animals within the focal group containing the tagged whale. While it is not possible to ensure that some behavioural responses occurred that were not detected, no observations of the kinds of very strong, overt responses identified as shut-down requirements were observed. Finally, no stranding of cetaceans was observed from the aerial surveys of large areas of the Tongue of the Ocean after either individual playbacks or the experiment as a whole. Although reactions to the sonar were observed in beaked whales (Tyack *et al*., 2011), reactions on the part of the delphinids were more difficult to distinguish from normal variability in behaviour. There were no indications from any of the playbacks that whales were injured or otherwise harmed by the signals played to them (Table 4). The beaked whale that showed the strongest reaction was the one tagged and exposed to playback of sonar and then killer whale sounds in 2007. This whale had an unusually long ascent after exposure, an unusually long interval between deep foraging dives after the killer whale playback, and a prolonged avoidance reaction (Tyack *et al*., 2011). Models of diving physiology suggest that none of the changes in dive profile in response to this sonar playback posed a risk to the subject (Kvadsheim *et al*., 2012). The tag monitoring the response fell off 10 hours after playback, while the whale was still engaged in an avoidance response (Allen *et al*., 2013), so it is not known when its behaviour ceased being disturbed by the playback. This kind of strong directed avoidance response may be used as an indicator of risk of stranding, but this whale was positively identified from photos when re-sighted in apparently good health in 2008, 2009, 2011, 2012 and 2013 (Bahamas Marine Mammal Organization, unpublished data).

DISCUSSION

The evidence presented here suggests that the controlled exposure experiment achieved its objectives of providing novel empirical information to inform management decisions without causing injury, harmful or permanent changes in behaviour to experimental subjects. The effects of acoustic exposure may depend on various contextual factors including source operation, deployment environment, and individual characteristics, such as age, sex, behaviour, social and motivational state of the exposed animals (Southall *et al*., 2007; Ellison *et al*., 2012). Given the available information at the time about these kinds of acoustic signals and the focal species, there was no *a priori* reason to assume at the start of this experiment that it could not have caused harm for some individuals. The historical stranding record reveals MFA sonar exercises using these specific kinds of signals that coincide with cetacean strandings, (Cox *et al*., 2006; Brownell *et al*., 2009; D'Amico *et al*., 2009). The evidence for a link between sonar exposure and stranding is strongest for atypical mass strandings of beaked whales (D'Amico *et al*., 2009, Filadelfo *et al*., 2009). However, there was insufficient information about what components of the sonar exposure led to these strandings. This study was designed to measure the parameters of sonar exposure required to elicit behavioural responses that were safe for the subjects but that could be used as indicators of risk. However, it was difficult to be completely certain that these experiments would not injure or strand cetaceans. The results of the study, both in the types of behavioural responses observed (directed, sustained avoidance – see Tyack *et al*., 2011) and in the lack of any observed extreme short-term responses that might pose a risk to diving physiology or long-term responses that might pose a risk of stranding suggest that it may require specific conditions (e.g. sustained transmissions following initial responses, multiple sound sources, particularly reverberant environments, high sound exposure levels) to elicit extreme responses that could lead to stranding.

It remains possible, although very unlikely, that the experiment led to some level of harm that remained undetected. The scale-based approach adopted to detect negative consequences was as comprehensive as resources and technology would allow. Animals that had been exposed were followed, to the extent possible, until their behaviour returned to normal. Behavioural responses were generally too subtle for significant changes to be observed based upon surface visual monitoring alone, even though they did occur (Tyack *et al*., 2011). It is also important to note that the observation effort for detecting and mitigating harm, even when supported by considerable technological capability from the AUTEC range, was considerably greater than the effort that was required as mitigation as a condition of authorisation of the research or that would be required during potentially damaging use of high intensity sound sources such as sonars, pile driving and seismic air guns. Consequently, this study also raises questions about the utility of current mitigation of the effects of high intensity sound sources in the ocean that rely exclusively on visual observers, especially to monitor for highly cryptic animals such as beaked whales. Rather, an integration of visual and acoustic monitoring approaches with a priori acoustic modelling and explicit mitigation and shut-down protocols is a more effective and responsible mitigation approach, especially for particularly sensitive species or important habitat areas (e.g. Nowacek *et al*., 2013).

Tyack *et al*. (2011) combined the results of experimental exposures to two beaked whale subjects with acoustic and satellite tag monitoring of responses of beaked whales to actual sonar exercises. A key approach was to use sophisticated tags to extract the maximum amount of acoustic dosage and behavioural response information from a small number of experiments in order to inform less detailed opportunistic observations. The dose-escalation protocol was designed to detect the minimum exposure required to elicit a response. The duration and level of exposure during experiments was reduced to the minimum required to elicit a silencing response in beaked whales that were using echolocation to forage. The duration and source level of exposure were markedly lower than those used during sonar exercises; the sound exposure protocol was designed specifically to minimise exposure to the minimum required to obtain the required scientific results. Combining results from experimental and opportunistic studies made it possible to reduce the number of animals exposed to sound that was transmitted as part of an experiment as opposed to ongoing sonar training. This kind of reduction in number of animals exposed and reduction in intensity of exposure forms a key component of animal welfare regulations and was endorsed by Huntingford (1984), and that for experimental exposures that could pose a risk to the subject.

This research was designed to help guide management of the risks from sonar exposure. The results from Tyack *et al*. (2011) suggested that responses consistent with elevating risk occurred at sound exposures of about 140dB, well under the previous regulatory thresholds. The threshold used by the US regulator to predict disturbance was subsequently changed to a 140dB step function threshold within several years of the publication of the Tyack *et al*. (2011) study (NMFS, 2013). If this change in threshold provides greater and more realistic protection of beaked whales from risks of exposure to levels that heretofore were thought to be safe, then the small costs to the experimental subjects needs to be weighed against the benefit to beaked whale populations worldwide.

The experiment complied with the guidelines suggested by Gales *et al.* (2009) and Huntingford (1984), overall, achieved an appropriate balance between the costs to the animals involved and the benefits in terms of novel data on reactions of animals to sonars. The results of this experiment and the mitigation measures used have directly served to inform subsequent research efforts involving sonar and marine mammals (e.g. Southall *et al.,* 2012; DeRuiter *et al.,* 2012; Goldbogen *et al.,* 2013; Miller *et al.,* 2012). None of these later studies exactly replicated the mitigation measures used here, because the situations and subjects involved were different and because results are increasingly showing that responses are likely to be subtler behavioural changes rather than physical injury or responses likely to lead to a stranding event. However, the basic approaches of an integrated system of visual and acoustic monitoring of the survey area (as well as use of sound propagation modelling in real time to visualise ranges of potential impacts) were derived to some extent from the Tyack *et al*. (2011) study. As described by Nowacek *et al*. (2013) for a multi-faceted monitoring and mitigation approach for a seismic survey conducted in critical feeding habitat for endangered western grey whales, the relatively costly and time-consuming mitigation process outlined here was beyond what was required and is likely beyond what may be possible in all subsequent studies. There is likely a justifiable reduction in certain elements from the broad approach described here, particularly in studies where such sounds are relatively common and species being tested have some baseline information on their basic behaviour and typical kinds of behavioural responses. A logical progression may be to retain certain fundamental protocols while relaxing others. Key elements that would logically be retained include sound propagation modelling to inform appropriate selection of exposure location, shut-down for animals within close proximity to loud sources and more protective protocols for particularly sensitive or endangered species. Other precautionary requirements such as aerial surveys for stranded animals following every sound transmission or shut-down of sound sources immediately based on cessation of sound production in animals in the vicinity could and have been have been relaxed based on scientific results occurring in subsequent studies. This is especially the case when considering best practices for smaller scale behavioural response studies, where budget limitations may in part dictate realistic protocols. Priority should be given to integrated mitigation measures for the specific circumstance of the study that ensure robust metrics to determine appropriate behavioural response data are collected while achieving mitigation goals.

In conclusion, this study found that a robust and open discussion of ethical issues associated with field experiments led to a mitigation protocol that allowed the meeting of scientific and applied objectives while minimising adverse impacts to the subjects of the study and animals nearby. As scientists develop more experience with novel kinds of study, their increased experience may support reduction of precautionary mitigation and monitoring measures, but each stage of the process requires careful and open evaluation of benefits and risks. This study agreed with Farnsworth and Rosovsky (1993) that scientists should include an explicit consideration of ethical issues in their peer-reviewed scientific publications, especially when there is uncertainty as to the impact of new study designs or when studies may have adverse or large scale impacts.

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