

A Study to Interpret the Biological Significance of Behavior Associated with 3S Experimental Sonar Exposures

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LONG-TERM GOALS

The long term goal of the 3S international cooperative research program is to investigate behavioral reactions of cetaceans to naval sonar and various control sounds, and the sound exposures required to elicit responses, in order to establish safety limits for sonar operations. 3S project efforts have been focused upon informing the ‘exposure-effect assessment’ component of a full risk-assessment framework (Fig. 1), which has been identified as a crucial missing component of the risk-assessment need (Boyd et al., 2008).

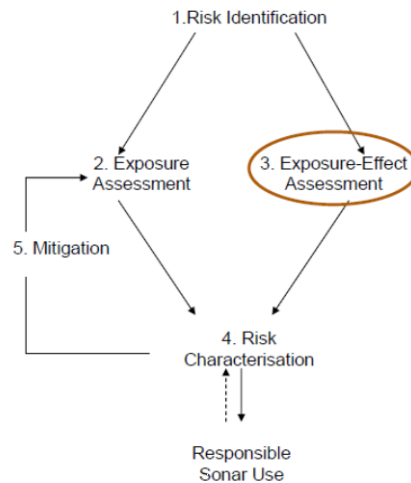


Figure 1. A risk-assessment framework. To characterize risk, both assessment of the amount of exposure in the environment and assessment of the relationship between exposure and effect (circled) are required.

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A significant challenge to establishing safety limits for sonar operations is in specifying what types of ‘effects’ need to be considered in the exposure-effect assessment. Under the US Marine Mammal Protection Act, both direct harm and harassment of marine mammals is regulated, so effects of concern cover a wide span of possible effects. Under new guidelines specified in the European Marine Strategy Directive, nations are required to specify ‘healthy state of the marine environment’, which could potentially include evaluation of a large array of potential effects from underwater noise emissions. Considerable recent focus has been shifted to understanding how behavioral effects might have longer-term consequences for individuals and populations of cetaceans (NRC, 2005). Modelling efforts under the PCAD working group have demonstrated the potential for behavioral effects to influence vital rates via energy balances (New et al., 2013). Behavioral effects of noise exposure may be shaped by anti-predator adaptations (Frid & Dill, 2002), and it is likely that natural selection will have operated substantially on the behavioral choices that underlie reactions to anthropogenic noise. The proposed research is therefore relevant to the US Navy to advance research in the area of determining ways of assessing ‘effects’ that have the specific potential to influence life history traits, and could therefore be considered harassment under the Marine Mammal Protection Act.

OBJECTIVES

The specific objectives of this project are: 1.) use of state-classification modelling (e.g. hidden Markov models, state-space modelling) to assess how sonar exposure might affect functional behavioral time budgets across 3S species; 2.) quantitative comparison of behavior, and behavioral changes, during sonar presentation and playback of killer whale sounds across the 3S species; and 3.) quantification of the possible impacts of sonar exposure on energy expenditure via linkage of respiration behavior and underwater activity recorded by Dtags.

APPROACH

The analyses in this study will take advantage of baseline data available for each target species, as well as the full range of experimental exposures conducted, including silent approaches as negative control and killer whale playback as positive control stimuli (Table 1).

Table 1. The 3S data-set collected to date.

| <u>Species</u> | <u># tagged</u> | <u># with CEE</u> | <u># of sonar exposures</u> | <u># of silent controls</u> | <u># of KW playbacks</u> |
|---|-----------------|-------------------|-----------------------------|-----------------------------|--------------------------|
| Killer whale (<i>O. orca</i>) | 10 | 4 | 8 | 1 | 2 |
| LF pilot whale (<i>G. melas</i>) | 30 | 8 | 14 | 4 | 8 |
| Sperm whale (<i>P. Macrocephalus</i>) | 10 | 4 | 10 | 2 | 5 |
| Humpback whale (<i>M. novaeangliae</i>) | 28 | 11 | 20 | 11 | 8 |
| Minke whale (<i>B. acutorostrata</i>) | 1 | 1 | 1 | 1 | 0 |
| N bottlenose whale (<i>H. ampullatus</i>) | 6 | 1 | 1 | 0 | 0 |
| TOTAL | 80 | 29 | 54 | 19 | 23 |

Technical approach for objective 1: Behavioral state modeling to derive time budgets

The goal of this task is to classify behavioral time series into functional behavioral states, enabling quantification of time budgets and proxies for costs and benefits of behavior within each functional state. Internal and external drivers of behavior may be combined by considering behavioral time series to arise from discrete functional units (‘functional states’) that are associated with the fulfillment of a

particular proximate or ultimate goal or set of goals based on a priori hypotheses (Nathan et al., 2008). States may be classified directly from data ('behavioral state'), such as area-restricted search, or refer to an underlying motivation that drives observed behavior ('motivational state', Bindra, 1978), such as hunger level. With advances in statistical computing, there is increasing scope to estimate these states within more realistic hidden process models that distinguish the observation and underlying ('hidden') process explicitly (Patterson et al., 2008; Schick et al., 2008) and integrate multivariate and multi-scale descriptors of behavior (McClintock *et al.*, 2013). A realistic approach is to allow behavioral states to vary over time (state-switching).

The framework we will apply will include analysis of a broad suite of recorded behaviors, and specifies the use of functional 'currencies' (e.g. feeding rates, locomotion costs) in conjunction with state-modelling to measure benefits and costs associated with a functional state (Fig. 1; Isojunno & Miller, in press). If functional currencies can be estimated given a state, fitness consequences can be evaluated with or without evidence of a specific behavioral response. This 'functional state approach' helps to frame the cross-disciplinary links between the motivating currency, proximate constraints and ultimate consequences of behavior, and encourages the view that behavioral context is a signal that could potentially fill in knowledge and data gaps of individual-based approaches to population consequence, rather than noise adding unexplained complexity to behavioral records.

The statistical analysis methodology for this objective will utilize state modelling (hidden or non-hidden) to quantitatively classify behavior into functional states determined for 3S study species (Jonsen et al., 2013). The time-series of functional behavioral state, and the quantitative indicators of benefits and costs within the state (feeding indicators, energy expenditure, social investment), are then used to test for systematic variations across different exposure conditions (tagging periods, baseline data periods, sonar-exposure sessions, killer-whale playbacks).

We will make use of this approach to quantitatively evaluate the effects of sonar exposure and killer whale playback of 3S species, starting with the long-finned pilot and humpback whale data sets. Currencies of cost/benefit are fairly easy to measure with these species. Cost can be quantified using ODBA or from breathing patterns (Objective 3). Benefits can be scored using deep dive buzzes and lunges as indicators of feeding in long-finned pilot and humpback whales, respectively. The primary challenge in applying the method to these other species is to successfully establish the functional behavior states employed by these species. The highly social nature of long-finned pilot whales may justify classification of states at both the group and individual level (Figure 6). Quantitative metrics of production of social sounds by this species (Objective 2), will aid in classifying group behavioral state, and may also serve as a benefit currency for this social species.

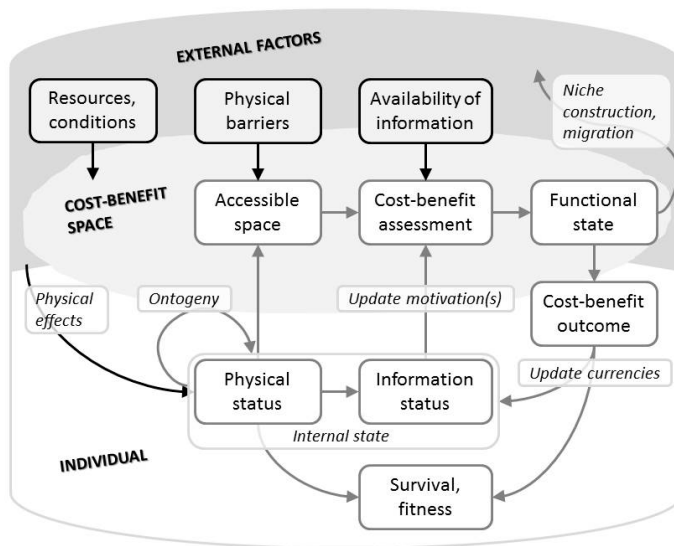


Figure 2. A framework for context-dependent behavior, inspired by Nathan et al. (2008). This ‘functional state approach’ consists of the focal individual, its biotic and abiotic environment (external factors), and their interface (cost-benefit space). Cost-benefit space is the outcome of behavioral options available to an individual, such as a trade-off between foraging and predation risk. Behavioral options are associated with a set of proximate goals or ultimate motivations (e.g. food, information). These options are limited physically and physiologically (accessible space) through individual history (ontogeny), current physical status (reproductive state, body condition and homeostasis) and physical barriers. Cost-benefit assessment reflects the individual’s internal mechanism for assessing a behavioral option. Information status encompasses cues, information and memory from both sensory and non-sensory inputs in the somatic nervous system. Functional state is the realized behavioral option exhibited by the animal and gives rise to a collection of behavioral traits that lead to a cost-benefit outcome. Currencies are the measures of the cost-benefit outcome of the functional state and feed back to the internal state of the individual, with effects on subsequent behavioral options, functional states and fitness (from: Isojunno & Miller, in press).

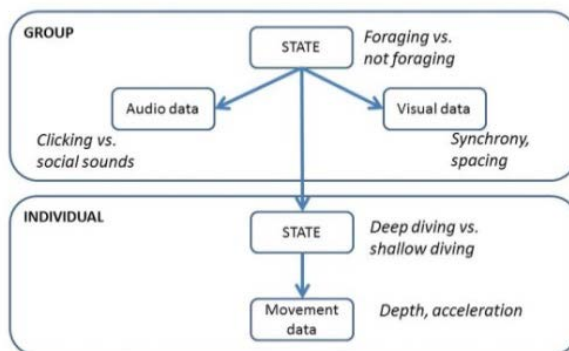


Figure 3. Initial schematic of nested social and individual state classification structure for long-finned pilot whales using social indicators on the group level, and tag data on the individual level.

Technical approach for objective 2: Contrasting response to sonar exposures and predator playbacks:

In this task, additional data and statistical analyses will be conducted to explicitly contrast the behavior of whale subjects in the 3S dataset during sonar exposure and playback of killer whale sounds, with the goal to evaluate the risk-disturbance hypothesis (Frid & Dill, 2002). Focused analysis of behavior changes following playback of killer whale sounds to sperm (Curé et al., 2013) and long-finned pilot whale (Curé et al., 2012) have been published, and response of humpback whales to killer whale playback is part of the current 3S² work plan (see Related Projects). While separate analyses of responses to killer whale playback and sonar exposure (Miller et al., 2012) provide a qualitative basis for contrasting behavioral changes during acoustic exposures, here we propose a targeted statistical contrast of both the intensity and direction of behavioral effects of these exposure types.

The statistical aspects of this task will be accomplished using the functional-state modeling approach of Task 1. Work under this task also will benefit from additional scoring of Dtag audio recordings to create quantitative indicators of social sound production by the 3S target species.

Technical approach for objective 3: Quantifying potential energetic impacts of sonar exposure:

The goal of this task is to evaluate possible energetic-expenditure changes due to sonar exposure, based upon linkages of underwater activity and breathing events recorded by Dtag sensors. We will use the fine-scale kinematic data recorded by Dtag to evaluate energetics and related breathing patterns in 3S study species. The aim will be to analyze: **1)** the strength of correlations between respiration rates and underwater activity levels (Williams et al., 2004; Williams and Noren, 2009); **2)** to what extent respiration timing (in addition to rate) alters the predicted oxygen taken up by each breath and therefore creates a more accurate predictor of metabolic requirements; and **3)** the impact of disturbance from naval sonar transmissions or playback of killer whales sounds on energetic requirements. Energetic requirements might increase due to behavioral changes such as increased swimming speed associated with avoidance (Miller et al., 2012). This analysis will enable direct energy expenditure contrasts by exposure condition, and will also provide useful methods to quantify the ‘cost’ component of behavior within different functional states (Task 1).

The framework for this analysis will be to build a time-series model of the predicted, ongoing oxygen store carried by each study animal:

$$O_2 \text{ store}_{n+1} = O_2 \text{ store}_n - O_2 \text{ costs}_n + O_2 \text{ uptake}_n \quad (\text{Eq. 1})$$

where, $O_2 \text{ store}_{n+1}$ and $O_2 \text{ store}_n$ are the total amount of O_2 in the body (including lungs, blood and muscle stores) at each time step. $O_2 \text{ costs}_n$ is the O_2 utilized for metabolic activity, and $O_2 \text{ uptake}_n$ is the O_2 acquired through breathing for each time step. $O_2 \text{ costs}_n$ will be modeled using estimates of basal metabolic rate plus predicted or empirical relationships derived from underwater activity metrics (Williams et al., 2004). Activity metrics will include speed (calculated through kinematic measurements and analyses of flow noise; Simon et al., 2012), thrusting movements or acceleration (ODBA, calculated by kinematic measurements; Wilson et al., 2006), and fluke stroke rate relating to thrusting movements (Goldbogen et al., 2006).

Respiration times will be extracted from time-depth records via surfacing cues, following published methods (Miller et al., 2010). The amount of oxygen acquired for each breath can be quantified as:

$$O_2 \text{ uptake}_{\text{breath}} = VT (IO_2 - XO_2), \quad (\text{Eq. 2})$$

where V_T is tidal lung volume or amount of liters air exchanged per breath ($L \text{ breath}^{-1}$) [12], IO_2 is the O_2 proportion of inhaled air, and XO_2 is the O_2 proportion of exhaled air.

A key advance in our proposed work is that O_2 exchanged per breath will vary based upon the modeled oxygen store, rather than a constant amount per breath assumed when breathing rate alone is used (Williams & Noren, 2009). It is well known that gas exchange in the lung will depend upon the relative partial pressures of the gas in the lung and the time available for the exchange to take place (Schmidt-Nielsen, 1997). While data for oxygen uptake is not known for killer whales, it is possible to model this process (e.g. Wilson et al., 2003; Fahlman et al., 2006).

Thus, the energetic-expenditure analysis will consist of two components: 1.) the estimate of O_2 use based upon basal metabolic rates and activity level, and 2.) the estimate of O_2 uptake per breath based upon breathing times and the current estimated oxygen score. Because this model accounts for underwater activity and variable oxygen uptake per breath, it is expected to be a more accurate representation of the metabolic rate than breathing rate alone. As actual metabolic rate was not measured directly during 3S experiments, a full external validation of the approach is not possible. Instead, we will validate the model by correlating predicted oxygen usage with activity levels.

WORK COMPLETED

Work under this project started on 01 July, 2014. Staff positions for the project have been filled, and work has begun on initial stages of the ‘functional state’ modelling approach (Objective 1), coordinating with the MOCHA project team (see Related Projects). The required acoustic analyses of tag recordings has begun (Objective 2), with an initial focus on the acoustic behaviour of long-finned pilot whales recorded during killer whale playbacks conducted in 2013 and 2014.

Graduate student M. Roos has implemented an initial version of the metabolic rate analysis from breathing times of killer whales (Objective 3). Roos presented her results in the 2014 Effects of Sound on Marine Mammals conference in Amsterdam and the 2014 Biologging Symposium in Strasbourgh.

RESULTS

For objective 3, graduate student M. Roos has made use of existing respiratory parameters for the killer whale (e.g. Kriete, 1995), to make an initial analysis of 3S killer whale Dtag data. In this initial oxygen model, oxygen use by the animal was calculated as basal metabolic rate plus a simple cost per fluke stroke performed by the tagged whale. Oxygen uptake per breath was modelled as a function of oxygen store at the time of each breath (Fig. 4).

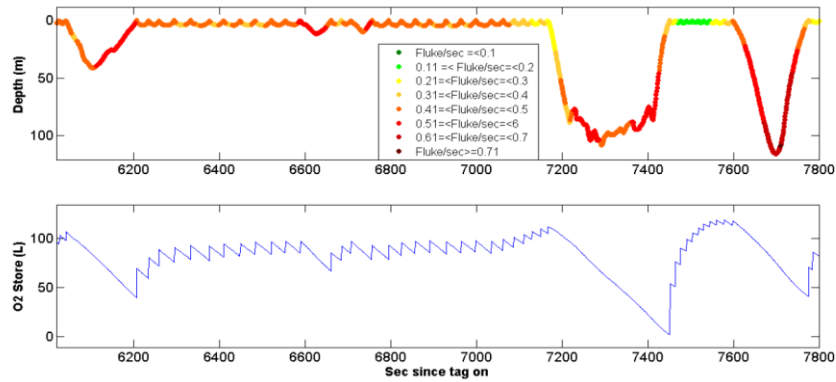


Figure 4. Data segment from tagged killer whale oo09_144a. Top: dive profile with fluking activity encoded as color. Middle: predicted O₂ store time series. Note that O₂ consumption tracks the activity level, while O₂ uptake at each breath varies with the O₂ store at the time of each breath.

Evaluations using this model across 12 tagged killer whales indicate much stronger correlations of activity and O₂ usage from our uptake modeling approach than breathing rate alone. Correlations of fluke stroke number versus oxygen uptake over 15 minute intervals for breathing rate were weak (r^2 ranged from 0.034-0.66), while much stronger relationships were found using the oxygen uptake model in which O₂ uptake varies by breath (r^2 ranged from 0.89-0.98; see example in Fig. 5).

Applying these new estimates of metabolic rate (O₂ use) to a sonar exposure experiment (Fig. 5) greatly improves our ability to robustly estimate metabolic rate changes due to increased activity associated with behavioral response (Miller et al., 2012). During the 15-min period of greatest activity for whale subject oo09_144b, breathing rate alone did not increase (Fig. 5, left panel). However, use of the model allowing O₂ uptake per breath to vary by oxygen store (Fig. 5, right panel inset box) indicated a greater metabolic rate during that period of high activity (arrow in Fig. 5, right panel). This strong difference arose because the animal strongly changed the timing of its breaths, with substantially longer inter-breath intervals during the high-activity period. Higher uptake of O₂ per breath is expected after longer apnea periods, a factor which is ignored when using breathing rate alone, but which is effectively captured by use of the O₂ uptake model.

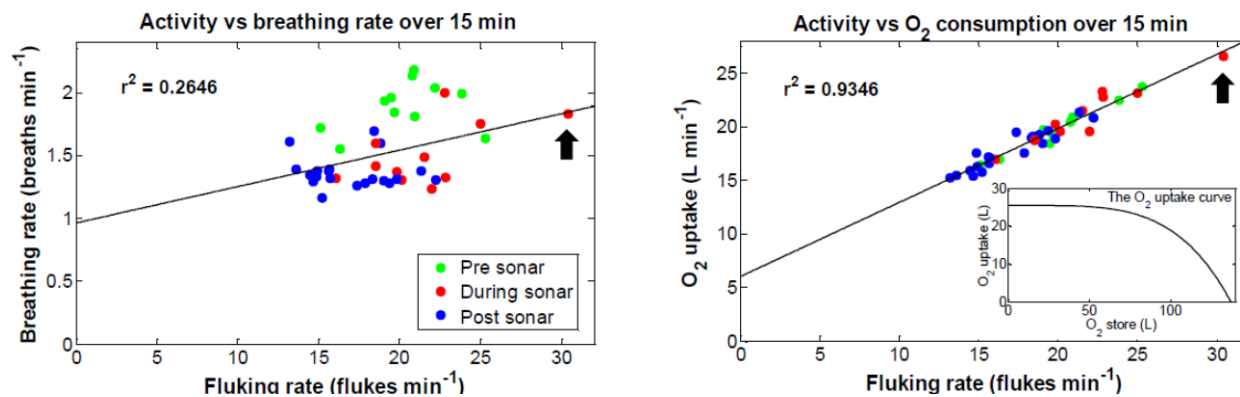


Figure 5. Scatter plot of the fluking rate versus oxygen uptake using breathing rate (left panel), and the full O₂ uptake model (right panel) for tagged whale oo09_144b. The O₂ uptake curve used in the oxygen model illustrated here is shown in the right panel inset box. Symbol colors indicate the phase of the sonar experiment for each 15-min interval. The arrows on the two panels indicate the identical 15 min period with high activity levels during an avoidance response by the tagged whale. Note much stronger correlations resulting from use of the uptake model which allows O₂ uptake to vary depending upon the O₂ store (right panel, inset box).

RELATED PROJECTS

This study is the third phase of the project “Cetaceans and naval sonar: behavioral response as a function of sonar frequency” award number N00014-08-1-0984, which expired in 2011. The second phase of the 3S project is “3S²: Behavioral response studies of cetaceans to navy sonar signals in Norwegian waters” which remains an ongoing project. Statistical support and collaboration is ongoing with the MOCHA project award N00014-12-1-0204. Collaborative research is pursued with Kelp Marine PI Fleur Visser under award N00014-11-1-0298. Additional support for this project is provided by French Ministry of Defence.

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