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Ronald A. Kastelein, Willem C. Verboom, Nancy Jennings, Dick de Haan, Sander van Der Heul. The influence of 70 and 120 kHz tonal signals on the behavior of harbor porpoises () in a floating pen. *Marine Environmental Research*, 2008, 66 (3), pp.319. 10.1016/j.marenvres.2008.05.005 . hal-00501970

HAL Id: hal-00501970

<https://hal.science/hal-00501970>

Submitted on 13 Jul 2010

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Accepted Manuscript

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PII: S0141-1136(08)00159-1
DOI: [10.1016/j.marenvres.2008.05.005](https://doi.org/10.1016/j.marenvres.2008.05.005)
Reference: MERE 3257

To appear in: *Marine Environmental Research*

Received Date: 6 August 2007
Revised Date: 3 May 2008
Accepted Date: 9 May 2008

Please cite this article as: Kastelein, R.A., Verboom, W.C., Jennings, N., de Haan, D., van der Heul, S., The influence of 70 and 120 kHz tonal signals on the behavior of harbor porpoises (*Phocoena phocoena*) in a floating pen, *Marine Environmental Research* (2008), doi: [10.1016/j.marenvres.2008.05.005](https://doi.org/10.1016/j.marenvres.2008.05.005)

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Twice revised ms MERE-D-07-00146. Sent to MER on 4 May 2008.

**The influence of 70 and 120 kHz tonal signals on the behavior of harbor porpoises
(*Phocoena phocoena*) in a floating pen**

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Abstract

Two harbor porpoises in a floating pen were subjected to five pure tone underwater signals of 70 or 120 kHz with different signal durations, amplitudes and duty cycles (% of time sound is produced). Some signals were continuous, others were intermittent (duty cycles varied between 8-100 %). The effect of each signal was judged by comparing the animals' surfacing locations and number of surfacings (*i.e.* number of respirations) during test periods with those during baseline periods. In all cases, both porpoises moved away from the sound source, but the effect of the signals on respiration rates was negligible. Pulsed 70 kHz signals with a Source Level (SL) of 137 dB had a similar effect to a continuous 70 kHz signal with a SL of 148 dB (re 1 μ Pa, rms). However, a pulsed 70 kHz signal with a SL of 147 dB had a much stronger deterring effect than a continuous 70 kHz signal with a similar SL. For pulsed 70 kHz signals (2 s pulse duration, 4 s pulse interval, SL 147 dB re 1 μ Pa, rms), the avoidance threshold Sound Pressure Level (SPL), in the context of the present study, was estimated to be around 130 dB (re 1 μ Pa, rms) for porpoise 064 and around 124 dB (re 1 μ Pa, rms) for porpoise 047. This study shows that ultrasonic pingers (\geq 70 kHz) can deter harbor porpoises. Such ultrasonic pingers have the advantage that they do not have a “dinner bell” effect on pinnipeds, and probably have no, or less, effect on other marine fauna, which are often sensitive to low frequency sounds.

Keywords: Acoustic alarm; Avoidance threshold level; Bycatch; Dolphin; Fisheries; Fishing; Nets; Odontocete; Pinger; Ultrasound.

1. Introduction

Many harbor porpoises (*Phocoena phocoena*) are accidentally caught in gillnets and drown (Trippel *et al.*, 1996; Read *et al.*, 2006). One potential alternative to reducing bycatch of small odontocetes by time and area closures of fisheries or by fish gear modifications (Read *et al.*, 2006), is to deter the animals from the nets acoustically. Harbor porpoises are deterred by certain anthropogenic underwater noises (Amundin and Amundin, 1973; Polacheck and Thorpe, 1990; Johnston, 2002; Olesiuk *et al.*, 2002; Koschinski *et al.*, 2003; Kastelein *et al.*, 2005a; Carstensen *et al.*, 2006; Madsen *et al.*, 2006).

Field studies with acoustic alarms on set gillnets have led to reduced harbor porpoise bycatch (Lien *et al.*, 1995; Kraus *et al.*, 1997; Laake *et al.*, 1998; Trippel *et al.*, 1999; Gearin *et al.*, 2000; Anonymous, 2000; Barlow and Cameron, 2003). Behavioral studies in the field show that porpoises avoid acoustic alarms (Laake *et al.*, 1998; Culik *et al.*, 2001; Johnston, 2002; Olesiuk *et al.*, 2002).

Acoustic alarms work by eliciting avoidance behavior and in some cases perhaps by alerting animals. One undesirable side-effect of alarms is noise pollution, which is increasingly believed to be damaging to the marine environment. Knowledge of the effects of alarm noises and of the hearing systems and behavior of many marine animals is very limited, but marine animals are likely to be disturbed by noise in their environment, and intense sounds may have negative physiological, auditory, and behavioral effects. For marine mammals, sound is particularly important as a means of communication, and echolocating mammals also use sound for orientation, and to locate prey, conspecifics and predators.

Therefore, sounds used in acoustic alarms should have characteristics such that they reduce odontocete bycatch, but also cause minimal noise pollution.

So far, the fundamental frequencies of signals of most commercially available acoustic alarms to reduce small cetacean bycatch have been below 20 kHz, although some recently developed pingers produce ultrasonic signals (Kastelein *et al.*, 2007). However, when not masked by background noise, harmonics produced by conventional alarms may be more audible to harbor porpoises than the fundamental frequency, as the hearing sensitivity of harbor porpoises increases up to around 120 kHz (Kastelein *et al.*, 2002).

The ideal alarm would be audible to harbor porpoises, but not to fish (Popper *et al.*, 2003) and pinnipeds (Southall *et al.*, 2007). The range of best hearing of harbor porpoises is 32-140 kHz (Kastelein *et al.*, 2002 – range highest sensitivity + 10 dB). Therefore, alarms designed to deter harbor porpoises should have their fundamental frequency between 32 and 140 kHz. If alarms are to be inaudible to pinnipeds, their frequency should be > 64 kHz (Southall *et al.*, 2007). Such ultrasonic signals have the advantages over audible signals that less energy is needed to produce them, and that they are somewhat more subject to attenuation over distance. Kastelein *et al.* (1997) showed that harbor porpoises can be deterred by ultrasound, but the extent of the exclusion zone caused by the signals used in that study is not clear. Culik *et al.* (2001) showed that harbor porpoises in the wild avoided an ultrasonic alarm producing sweeps between 20 and 160 kHz. Teilmann *et al.* (2006) tested three 200 ms signals between 100 and 140 kHz, with a signal interval of 4 s, and noticed displacement in porpoises in a pool, but the animals' reaction decreased over time. Kastelein *et al.* (2008) subjected a harbor porpoise to a continuous 50 kHz pure tone signal. The animal swam away from the sound source, and the avoidance threshold Sound Pressure level (SPL) was estimated to be around 108 dB (re 1 μ Pa, rms). The aim of the present study is to discover whether 70

and 120 kHz tonal signals are suitable for use in acoustic alarms to deter harbor porpoises, and if so, what sound parameters are important in causing the deterring effect.

2. Materials and methods

2.1. Study animals

The study was carried out on two male harbor porpoises that had been stranded and rehabilitated in the Netherlands. One animal (047) had stranded on the Dutch coast at the age of seven months, the other (PpSH064) on the German coast at the age of nine months (ages estimated from their body length and assuming that they were born in June). At the time of the study they were five and two years old respectively, healthy, weighed approximately 35 and 25 kg and were about 135 and 118 cm long respectively. The animals were fed fish five times a day: at 0845, 1030, 1230, 1430, and 1630 h.

2.2. Study area

The harbor porpoises were housed in a floating pen (34 m x 20 m; 2.4 m deep at the sides and 2.8 m deep in the center, containing two smaller pens; **Fig. 1**). The lower 10 cm of the surrounding pontoons (plywood boxes filled with Styrofoam and coated with fiberglass) were submerged. The bottom panel of the net was made of nylon and the four side panels were made of polypropylene; both types of net had a stretched mesh size of 9 cm and a twine diameter of 3 mm. The net was covered with algae. Seawater could flow through the pen, as could small fish and invertebrates, such as jellyfish.

The pen was in a harbor in the southwest Netherlands, at Neeltje Jans ($51^{\circ} 37' N$, $03^{\circ} 40' E$). The harbor is horseshoe-shaped (500 m x 280 m) with the entrance towards the north-east and near the middle of the leeward side of the Oosterschelde surge barrier. The barrier is only closed during exceptionally high tides and storms, and was open at the time of the study. Therefore the tidal amplitude inside the harbor was similar to that in the nearby North Sea (2.5 m). No shipping occurred within 2 km of the study area during the study. The depth of the harbor around the pen depended on the tide (**Fig. 1**). The sea floor below the pen was smooth and covered with sandy silt; salinity was around 34 ‰. During the study the underwater visibility (determined by Secchi disc reading) was between 1.0 and 3.1 m, and the average monthly water temperatures were 18 °C (July), 21 °C (August), 16 °C (September) and 13°C (October). During the experiments both animals were kept in the main pen (**Fig. 1**).

2.3. Underwater background noise

Background noise in the pen, mostly from wind and tide, was measured several times under conditions similar to those during which actual sessions were carried out. At high tide the swell caused squeaking, impulsive noises from the steel joints of pontoon elements of the floating pen, especially in north to north-east winds. Drainage noise occurred when water fell from the quayside into the harbor at falling tide. Background noise levels were determined in the range 100 Hz - 80 kHz and were converted to 'spectrum level' (dB re 1 $\mu\text{Pa}/\sqrt{\text{Hz}}$; **Fig. 2**).

Equipment used to measure the background noise consisted of a broadband hydrophone (Brüel & Kjaer 8101, 0-100 kHz), a voltage amplifier system (TNO TPD, 0-300 kHz) and an analyzer system (Hewlett Packard 3565, controlled by a notebook computer; sample frequency 260 kHz, $df = 31$ Hz; the result was presented in 1/3-octave bandwidths). The system was calibrated with a pistonphone (Brüel & Kjaer 4223) and a white noise 'insert

voltage' signal into the hydrophone pre-amplifier. Measurements were corrected for the frequency sensitivity of the hydrophone and the frequency response of the measurement equipment.

2.4. Stimuli

The porpoises were exposed to five ultrasonic signals (**Table 1**). The experimental electronics used to generate and project signals 1 and 2 were manufactured by Airmar Technology Corporation, Milford, U.S.A. Signals 3-5 were produced by a signal generator (Hewlett Packard, model 33120A). The high-frequency transducer, which was used for all five signals, was fixed in a standard Airmar gillnet pinger housing. To allow remote activation of the transducer, the internal battery (D-cell, 1.5 V) was replaced by an external power supply producing 1.5 V at the transducer end of the cable. The sound projection pattern of the transducer was omni-directional in the horizontal plane, and slightly flattened in the vertical direction due to an air cavity at the top of the transducer. The yellow polyurethane housing of the transducer had a diameter of 5 cm, was 15 cm long and weighed 0.3 kg. A 0.4 kg piece of lead, hanging from a string below the transducer, kept it vertically stable at 1.5 m below the water surface in the middle of the north side of the pen (**Fig. 1**).

The 70 kHz signal frequency was chosen to be above the hearing frequency range of pinnipeds, and the 120 kHz signal was chosen to coincide with the peak frequency of harbor porpoises' echolocation clicks (Møhl and Andersen, 1973; Verboom and Kastelein, 1995, 1997, 2003) and their most sensitive hearing (Kastelein *et al.*, 2002). For each signal (frequency — pulse duration — pulse interval combination) the Source Level (SL) used in the experiment was determined during a pre-test by increasing the amplitude until the porpoises swam away from the sound source.

2.5. Acoustic recording and analysis

The SPL distribution of the signals in the pen was measured by using a hydrophone (Brüel & Kjaer, model 8101) and a 30-m extension cable (Brüel & Kjaer) connected to a conditioning amplifier (Brüel & Kjaer, Nexus type 2690). The amplifier output was connected via a coaxial module (National Instruments, BNC-2090) to a computer (Joheco, PIV-1.6 GHz) with a data acquisition card (National Instruments, model PCI-MIO-16E-1), on which signals were digitized with 12 bits resolution. Sound signals were measured with a scan rate of 512 kHz. Monitoring, recording and analysis of signals were accomplished by the use of software modules developed by IMARES (National Instruments, Labview 6.0). A pistonphone (Brüel & Kjaer, model 4223) was used as a reference to calibrate the hydrophone, the conditioning amplifier and the analogue-to-digital conversion. Reference data were logged separately and used in the analysis module to calculate the SPL. The single frequency Fast Fourier Transform (FFT) analysis covered a bandwidth of 20 Hz to 256 kHz. The signals were subjected to FFT with Hanning window over the activated duration, and pure tones were analyzed in time blocks of 0.2 s (102564 samples).

All SPLs reported are corrected for the response curves of the hydrophone, the amplifier, and analogue-to-digital conversion. The SPL distribution was determined at 13 measurement locations in the pen with the hydrophone 1.5 m below the water surface, the same depth as the sound sources (**Fig. 3**).

The SL of each signal was calculated from the measured SPL at a distance of 2 m from the sound source by adding 6 dB (**Fig. 3**). Because it is not clear which sound parameter(s) influenced the porpoises' behavior, the SLs of the five signals are presented in three ways: 1) the SL based on the root-mean-square (rms) value of the signal amplitude; 2) the rms SL

averaged over the pulse interval (thus including the influence of duty cycle; 3) the source energy level (signal level x signal duration; **Table 1**).

2.6. *Experimental procedures*

The transducer was put into position (**Fig. 1**) several hours before a session started. A session was composed of a 15-minute baseline period (no acoustic stimulus produced), followed immediately by a 15-minute test period (stimulus produced). Daily, one session was conducted between 15.30 and 16.00 h. At this time of day the sun was behind the observers, so that their view of the animals was optimal. The observers were trained to recognize the individual porpoises. During the tests, people were not allowed on the pontoons.

An underwater listening system was used to check that the signals were produced during the test sessions. The system was composed of a hydrophone (LabForce 1 BV, model 90.02.01) and a heterodyne bat detector (Batbox III, Stag Electronics Steyning, UK). The hydrophone was placed 1.5 m below the water surface, 1 m away from the transducer (**Fig. 1**).

The five signals were tested between July and October 2002. A maximum of seven sessions per week were conducted. Tests were not carried out during rainfall or when wind speeds were above 4 Beaufort.

Initially the aim was to investigate the effect of duty cycle, and only 70 kHz Signals 1 (0.3 s) and 2 (1s) were considered. These two signals were tested 15 times each, in random order, between 2 July and 9 August 2002. Because less effect was seen than expected with 70 kHz Signals 1 and 2, two additional signals (Signals 3 (70 kHz) and 5 (120 kHz)) were tested with higher amplitudes. These signals were tested in random order between 11 August and 25 September 2002. Each signal was tested 15 times, once a day. To study the effect of duty cycle on the behavior of the porpoises, 70 kHz Signal 4 (2 s duration, 4 s interval, SL 147 dB)

was tested between 30 September and 23 October 2002. This signal was tested five times, instead of the intended 15 times, because due to poor weather conditions the number of days available was less than expected (the study period was limited by the end of the operational season for the floating pen).

2.7. Response variables and behavioral data recording

To quantify and compare the effects of the signals, we measured the displacement of the porpoises and changes in their breathing rate (as an indicator of their level of agitation). In order to do this, during the baseline and test periods, the locations where each animal surfaced were plotted on a map of the pen by two observers. Depending on the tide, the observers were 2 to 4.5 m above water level, on land to the west of the pen (**Fig. 1**). To facilitate the mapping of the positions, two ropes were strung across the pen (80 cm above the water level) with markers in the middle. On most days, the animals could be seen up to 1 m below the water surface, but on some days the water was so clear that the observers could see the bottom of the pen. Because on those days the porpoises were never seen swimming far away from their surfacing locations, it was assumed that these locations indicated their general swimming area. From the maps, the following response variables were derived: the number of respirations (or surfacings), and the distances between the transducer and the surfacings, during baseline and test periods.

2.8. Statistical analysis

Statistical analysis was done on Minitab release 14 (Ryan and Joiner, 1994) with a significance level of 0.05.

In preliminary analysis to investigate the effect of each signal on the distance of surfacings from the transducer, a separate analysis of covariance (ANCOVA) was used for each animal and for each signal, so that a total of ten ANCOVAs were carried out. In all, the session number was introduced as a covariate, so that any effect of session number could be both quantified and corrected for before the analysis of the only factor in the ANCOVA, which was the period (baseline or test). The assumptions of ANCOVA (normality of residuals and homogeneity of variances, see Zar, 1999) were not always conformed to exactly, but ANCOVA is robust to small departures from these assumptions (Zar, 1999).

To evaluate potential differences between the responses to the signals, further ANCOVAs were carried out in which the dependent variable was the difference between the distance from the transducer of surfacings in baseline and test (calculated for each session as the mean distance from the transducer in the baseline period, minus the mean in the test period). As above, the session number was included as a covariate; the only factor was the signal. One ANCOVA was carried out for each animal.

To investigate the effects of the signals on the number of surfacings in each session, a paired t-test was carried out for each signal and for each animal. When assumptions of the t-test were not met, a Wilcoxon matched pairs test was used as a substitute. In both cases the difference between the number of surfacings in baseline and test sessions were compared to zero, thus controlling for any effects of session number.

3. Results

Both animals moved away from the transducer when each of the five signals was emitted (**Tables 2 and 3, Fig. 4**). Animal 064 swam a similar distance away from the transducer in response to all five signals, but animal 047 responded significantly more

strongly to Signal 4 (70 kHz, 2 s pulse duration, 4 s pulse interval, SL 147 dB re 1 μ Pa, rms) than to the other signals (**Table 4 and Fig. 4**). A rough indication of the SPL in the general area occupied by each porpoise during test sessions with each of the five signals is shown in **Table 1**. For Signal 4 (70 kHz, 2 s pulse duration, 4 s pulse interval, SL 147 dB re 1 μ Pa, rms), the avoidance threshold SPL (the SPL at the boundary between the areas that animals generally occupied during the transmission of sounds and areas that they generally did not enter during transmission) was estimated to be around 130 dB (re 1 μ Pa, rms) for porpoise 064 and around 124 dB (re 1 μ Pa, rms) for animal 047.

In most cases, the number of surfacings was similar in baseline and test sessions (**Table 5 and Fig. 5**).

4. Discussion and conclusions

4.1. Reasons for the different effects of the five signals

Before and during the entire study period, the animals had a strong tendency to occupy the north-western corner of the pen. This was probably because the gangway to the pen was there (**Fig. 1**). From the gangway, the trainers arrived five times per day to feed the animals, and often started as soon as they stepped onto the pontoons around the pen. Therefore the transducer was placed at the northern end of the pen, so that the deterring effect of the test signals could be quantified. All five signals deterred the animals from the sound source, but only 70 kHz Signal 4 (2 s duration, 4 s interval, SL 147 dB) had an effect comparable to that of sonic pingers tested in previous studies with harbor porpoises in the same floating pen (Kastelein *et al.*, 1995; 1997, 2000, 2001). During test periods with Signal 4, the porpoises hardly visited their preferred north-western corner of the pen (**Fig. 1**).

The animals did not swim to the areas with the lowest SPLs, but tolerated higher SPLs (**Table 1**). Each animal had a preferred individual general surfacing area, suggesting that the porpoises had little influence on each other's swimming behavior. No effect of the duty cycle, when comparing pulsed signals of 70 kHz and the same SL (137 dB re 1 μ Pa, rms), was found (Signals 1 (0.3 s) and 2 (1 s)). For the 70 kHz pulsed signals (Signals 1, 2 and 4) the porpoises generally swam in an area with exposure SPLs of around 117 dB, while they accepted much higher exposure SPLs during continuous 70 kHz Signal 3 (129-131 dB; **Table 1**). More specifically, they swam further away from 70 kHz Signal 4 (2 s signal, 4 s interval, SL 147 dB) than from 70 kHz Signal 3 (continuous, SL 148 dB), although both were 70 kHz signals with similar source levels. Commercially available pingers are designed to produce pulsed signals mainly to reduce energy demands on the batteries, but the present study suggests that pulsation also increases the deterrent effect. Alarm devices for humans usually also consist of pulsed signals, as the perceived urgency is higher for pulsed signals than for continuous signals, to which habituation occurs also sooner (Edworthy *et al.*, 1991; Haas and Casali, 1995; Edworthy and Hellier, 2000).

The sensation level of a signal depends on its frequency, the porpoise's hearing threshold at that frequency (or frequency band), and the ambient noise level and spectrum. The harbor porpoise has a U-shaped audiogram (Kastelein *et al.*, 2002), and is more sensitive to 120 kHz signals than to 70 kHz signals. This probably explains why Signal 5 (120 kHz) had a relatively strong deterring effect, despite it being continuous and of lower SL than the 70 kHz signals.

During the present study, the signal detection level for part of the hearing frequency spectrum was influenced by the ambient noise at the study site. The theoretical masked detection thresholds (MDTs) for the 70 and 120 kHz test signals can be calculated as follows: $MDT = L_{sp} + CR - DI$, in which the MDT is for pure tones - dB re 1 μ Pa, L_{sp} is the

(equivalent) spectrum level of the background noise (in dB re $1 \mu\text{Pa}/\sqrt{\text{Hz}}$), CR the critical ratio of the hearing system (in dB re $\sqrt{\text{Hz}}$), and DI the receiving directivity index (in dB). The DI of harbor porpoises for 70 kHz is approximately 8.5 dB and for 120 kHz approximately 11.2 dB (based on a regression line through three data points; Kastelein *et al.*, 2005b). No information is available on the CR of harbor porpoises, but it can be estimated from human data and the measured CR of bottlenose dolphins, *Tursiops truncatus* (Johnson, 1968; estimated CRs: 45.5 dB at 70 kHz and 49 dB for 120 kHz). Due to the spectrum and level of the ambient noise in the floating pen, the theoretical masked hearing thresholds for the two test frequencies in the present study were similar (76 and 74 dB; **Table 1**). This calculation shows that all five test signals were audible to the porpoises throughout the floating pen.

The porpoises tolerated lower exposure SPLs for 120 kHz signals than for 70 kHz signals. This is expected considering that they have higher hearing sensitivity for 120 kHz signals than for 70 kHz signals (**Table 1**). However, the theoretical masked detection thresholds (hearing thresholds determined by the background noise level) are similar for both test frequencies. It is possible that the 120 kHz continuous signal interfered with the porpoises' echolocation and was thus tolerated only at lower levels, or that harbor porpoise critical ratios around 120 kHz are lower than expected based on extrapolating those of bottlenose dolphins.

4.2. Effects on other marine fauna

Ultrasonic pingers > 70 kHz are not audible to pinnipeds, so have the advantage that they cannot attract pinnipeds to the catch in gill nets (by the so-called “dinner bell” effect).

The hearing frequency range of most fish is up to 2 kHz (higher for clupeid fish; Popper *et al.*, 2003). The higher the frequency of a pinger, the less likely it is that fish species will

react to it. Some commercially available pingers produce, in addition to the advertised signals, low frequency signals to which some fish species react behaviorally (Kastelein *et al.*, 2007). Such reactions may lead to reduced catch rates, which may encourage fishermen not to use the pingers.

4.3. Limitations

Some caution is needed when interpreting the data presented here. Only two animals were available for the present study, so it may be risky to extrapolate the findings to all members of the species. Age, sex, location, context and experience may influence the behavior of individuals, and differences in the reactions of two harbor porpoises to pingers were described by Kastelein *et al.* (2001). In addition, the animals in the present study may have influenced one another, although this is not likely because they occupied different areas in the pen, and did respond differently to the five test signals.

In the present study the alarm signals were switched on at the SLs reported. This sudden onset may have startled the porpoises. In the wild, a marine mammal can swim freely towards a sound source, and may be able to regulate the speed with which the exposure SPL increases. However, in the present study the porpoises did not habituate to the sounds during the 15-minute test sessions or over the course of the study (15 sessions over approximately 1 month per signal), and in a similar study porpoises returned to baseline behavior immediately after sounds ceased (Kastelein *et al.*, 2000). This suggests that little of the behavior of the animals observed during the 15-minute test sessions can be ascribed to a 'startle' effect.

The present study was limited by animal welfare considerations. The SL of the test signals was increased only to the point that the animals moved away from their preferred north-western corner of the pen, and not so high that they moved to the other side of the pen.

Also, the animals were not exposed to the alarm signals for very long time periods (*e.g.* for several days) to test potential habituation to the sounds.

Each odontocete species probably reacts differently to particular sounds. In a study in which a striped dolphin (*Stenella coeruleoalba*) and a harbor porpoise were subjected to the same alarm, the porpoise swam as far away as possible from the sound source, whereas the striped dolphin showed no behavioral response (Kastelein *et al.*, 2006). The results of the present study are probably only valid for harbor porpoises under the test conditions.

Nevertheless, the results are useful for evaluating which parameters are most likely to be effective in the field.

4.4. Suggestions for future research

The present study has shown that ultrasonic signals can deter harbor porpoises from a particular area for a certain period of time. However, the following research should be carried out to make acoustic alarms more efficient and less acoustically polluting:

- 1) Discover which parameters of sounds (*e.g.* pulse duration, duty cycle, SL, harmonic distortion, spectrum) are important in rendering them suitable for use as acoustic alarms. For this research, the SL must be more tightly controlled experimentally than was possible in this study.
- 2) Study the effects of masking on sounds. During rainfall, part of an alarm's spectrum may be masked by ambient noise. Observations over seven years of pinger research suggest that porpoises are less deterred by pingers during strong winds (which cause waves) and rainfall than during calm, quiet weather conditions (Kastelein *pers. obs.*).
- 3) Test the effect of potential pinger sounds on other marine fauna, such as fish, and on non-target marine mammal species.

- 4) Study potential habituation by porpoises to acoustic alarm signals.
- 5) Determine the optimal SL of alarms and their configuration in the nets (numbers of alarms needed, inter-alarm distances). For one alarm type this has already been studied (Larsen and Krog, 2007).

Acknowledgements

We thank Rob Triesscheijn, Mariska Muijsers, Natascha Lor, Brigitte Slingerland, Marjolijn Essenburg, Famke de Hond, Betty Braaksma and Paul Wensveen. Steven Christensen (Airmar Technology Corporation, Milford, NH, U.S.A.) provided the experimental Airmar alarm signal generating system. We thank Lee Miller (Odense University, Denmark), Finn Larsen (Technical University of Denmark), Simon Northridge (Sea Mammal Research Unit, St. Andrews, UK) and two anonymous reviewers for their comments on the manuscript. We thank Jan de Kam and Hans Jager (both of RWS, Topshuis, Neeltje Jans) for postponing dredging activities at Neeltje Jans for this study. The study was funded by SEAMARCO, Harderwijk, The Netherlands, and The Netherlands North Sea Directorate (contact: Christoph Reuther), and conducted under authorization of the Netherlands Ministry of Agriculture, Nature Management and Fisheries, Department of Nature Management, with endangered Species Permit FEF27 06/2/98/0184.

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Tables:

Table 1

Parameters of the five test signals used, Source level (SL, expressed in three ways), Sound Pressure Level (SPL) range (dB re 1 μ Pa, rms) in the pen, approximate SPL in the general area occupied by each porpoise during test sessions, and the theoretical masked detection threshold (MDT) of the signals for porpoises in the ambient noise conditions of the study area.

Sig- nal	n	Funda- mental frequency (kHz)	Pulse duration (s)	Pulse interval (s)	Duty cycle (%)	SL _{fund} (dB)	SL _{av} (dB)	SL _E (dB)	SPL range in pen (dB)	SPL in area occupied by porpoises (dB)	MDT (dB)
1	15	70	0.3	4	8	137	126	132	124-105	~115	76
2	15	70	1	4	25	137	129	135	124-105	~115	76
3	15	70	Cont.	0	100	148	148	--	137-118	~130	76
4	5	70	2	4	50	147	144	150	137-115	~120	76
5	15	120	Cont.	0	100	134	134	--	119-102	~110	74

n = number of sessions conducted

SL_{fund} = Source Level of fundamental frequency (rms pulse level) in dB re 1 μ Pa/1 m

SL_{av} = averaged Source Level per pulse cycle (including duty cycle influence) in dB re 1 μ Pa/1 m

SL_E = source energy level in dB re 1 μ Pa² s/1 m

Table 2

ANCOVAs (one for each signal) for the effect of period (baseline and test) with session as the covariate on the distance between surfacings and the transducer for animal 047; Df = degrees of freedom, Adj. MS = adjusted mean squares, F = test statistic, *P* = probability. Both animals moved away from the transducer when each of the five signals was emitted.

Source of variation	Df	Adj. MS	F	<i>P</i>
Signal 1				
Period	1	1849.2	90.57	0.000
Session (covariate)	1	247.2	12.11	0.001
Error	1281	20.4		
Total	1283			
Signal 2				
Period	1	2537.1	111.14	0.000
Session (covariate)	1	952.4	41.72	0.000
Error	1260	22.8		
Total	1262			
Signal 3				
Period	1	5388.0	336.98	0.000
Session (covariate)	1	786.5	49.19	0.000
Error	1257	16.0		
Total	1259			
Signal 4				
Period	1	18275.2	894.18	0.000

Session (covariate)	1	243.9	11.94	0.001
Error	405	20.4		
Total	407			
Signal 5				
Period	1	8854.8	379.57	0.000
Session (covariate)	1	539.9	23.15	0.000
Error	1296	23.3		
Total	1298			

Table 3

ANCOVAs (one for each signal) for the effect of period (baseline and test) with session as the covariate on the distance between surfacings and the transducer for animal 064; abbreviations as for **Table 2**. Both animals moved away from the transducer when each of the five signals was emitted.

Source of variation	Df	Adj. MS	F	P
Signal 1				
Period	1	294.76	16.93	0.000
Session (covariate)	1	376.19	21.61	0.000
Error	1395	17.41		
Total	1397			
Signal 2				
Period	1	638.5	32.35	0.000
Session (covariate)	1	2162.1	109.55	0.000
Error	1374	19.7		
Total	1376			
Signal 3				
Period	1	522.65	40.30	0.000
Session (covariate)	1	15.67	1.21	0.272
Error	1399	12.97		
Total	1401			
Signal 4				
Period	1	2107.3	66.55	0.000

Session (covariate)	1	244.6	7.72	0.006
Error	436	31.7		
Total	438			
Signal 5				
Period	1	3223.0	192.11	0.000
Session (covariate)	1	1269.5	75.67	0.000
Error	1482	16.8		
Total	1484			

Table 4

ANCOVAs (one for each animal) for the effect of signal with session as the covariate on the difference (baseline – test) in session mean distance between surfacings and the transducer; abbreviations as for **Table 2**. Animal 064 swam a similar distance away from the transducer in response to all five signals, but animal 047 responded significantly more strongly to Signal 4 than to the other signals.

Source of variation	Df	Adj. MS	F	P
Animal 047				
Signal	4	81.42	8.95	0.000
Session (covariate)	1	79.19	8.70	0.005
Error	59	9.10		
Total	64			
Animal 064				
Signal	4	12.27	1.86	0.129

Session (covariate)	1	43.85	6.65	0.012
Error	60	6.60		
Total	65			

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Table 5

Results of paired t-tests (where appropriate; otherwise Wilcoxon matched pair tests, marked with *) to compare the difference between numbers of surfacings in baseline and test (baseline minus test) to zero. Significant differences were found in only three cases. n = sample size (number of sessions), T/W = test statistic for paired t-test or Wilcoxon matched pair test, *P* = probability.

Animal 047	n	T/W	<i>P</i>
Signal 1	15	-0.29	0.778
Signal 2	15	-2.38	0.032
Signal 3	15	-1.29	0.217
Signal 4	5	0.0*	1.000
Signal 5	15	1.04	0.314
Animal 064			
Signal 1	15	2.34	0.035
Signal 2	15	1.25	0.233
Signal 3	15	-2.85	0.013
Signal 4	5	6.0*	0.787
Signal 5	15	-0.30	0.765

Legends to figures:

Fig. 1. Top view of the study area, showing the locations of the underwater transducer, hydrophone, equipment cabin with operator, the two observers, and the average water depths at the corners of the floating pen at high tide (HT) and low tide (LT).

Fig. 2. Background noise limits in the floating pen between 100 Hz and 100 kHz. Each measurement was done during a 32-60 s period. The system noise did not influence the acoustic measurements. Results are given as equivalent spectrum levels in dB re 1 $\mu\text{Pa}/\sqrt{\text{Hz}}$, at 1/3-octave centre frequencies.

Fig. 3. The Sound Pressure Level (SPL in dB re 1 μPa , rms) distribution in the floating pen for the five ultrasonic signals. SL = Source Level. Each column from top to bottom: Signals 1 and 2, Signal 3, Signal 4, and Signal 5 (see upper left hand corner column).

Fig. 4. The adjusted mean distances between the porpoises' surfacings and the transducer during baseline (white columns) and test (grey columns) periods with the five test signals ($n = 15$ sessions for Signals 1, 2, 3 and 5, and five sessions for Signal 4). A) Porpoise 047, and B) Porpoise 064. The bars indicate standard errors. For both animals, for all five signals, values for test periods are significantly higher than those for baseline periods.

Fig. 5. The adjusted mean number of surfacings of the porpoises during baseline (white columns) and test (grey columns) periods with the five test signals ($n = 15$ sessions for Signals 1, 2, 3 and 5, and five sessions for Signal 4). A) Porpoise 047, and B) Porpoise 064.

The bars indicate standard errors. An * indicates a significant difference between mean baseline and mean test values.

Figure 1

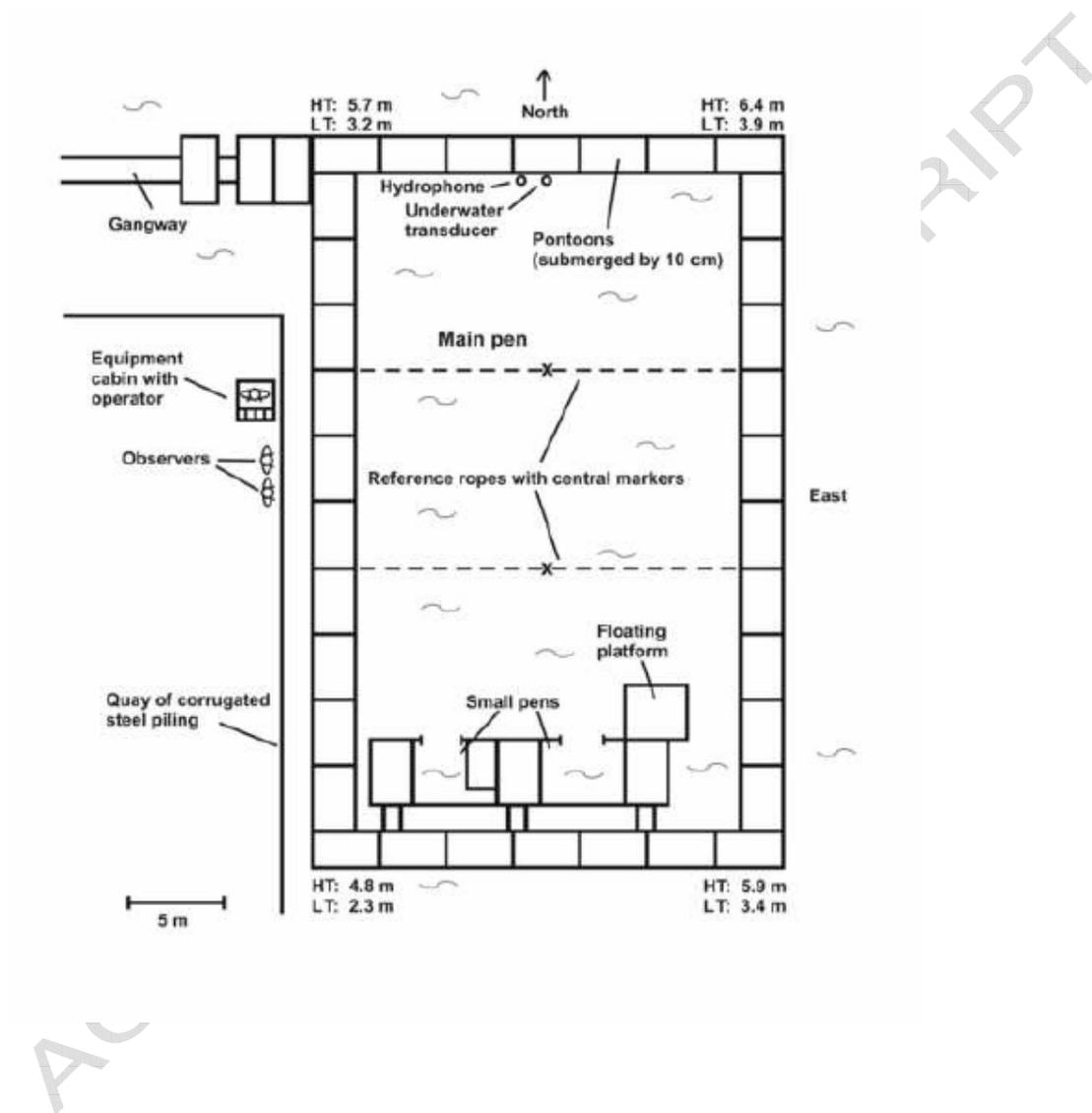
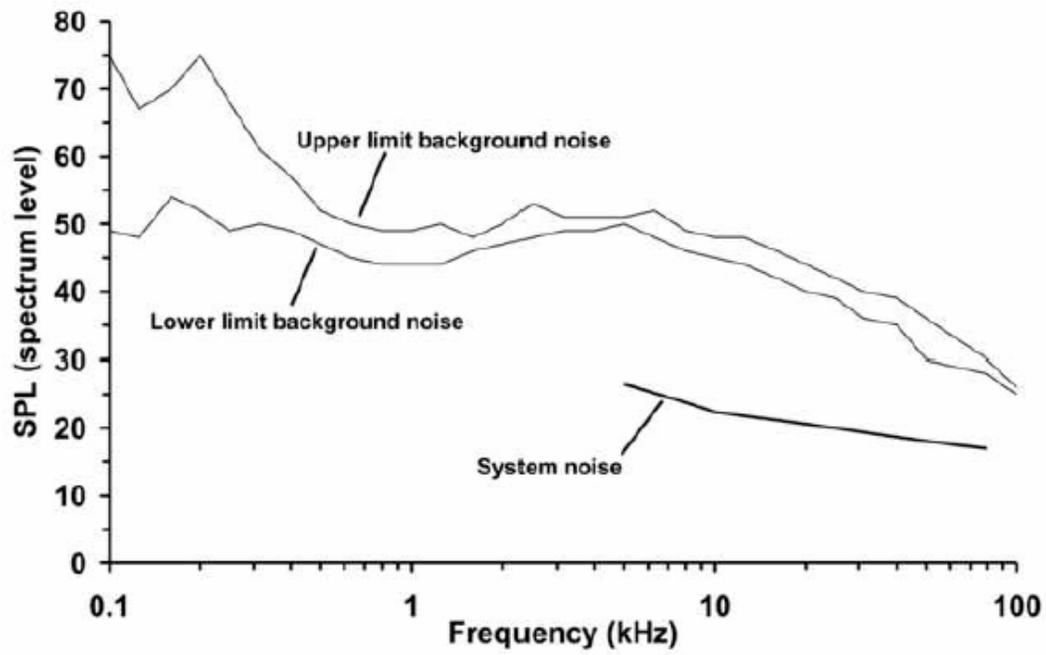


Figure 2



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Figure 3

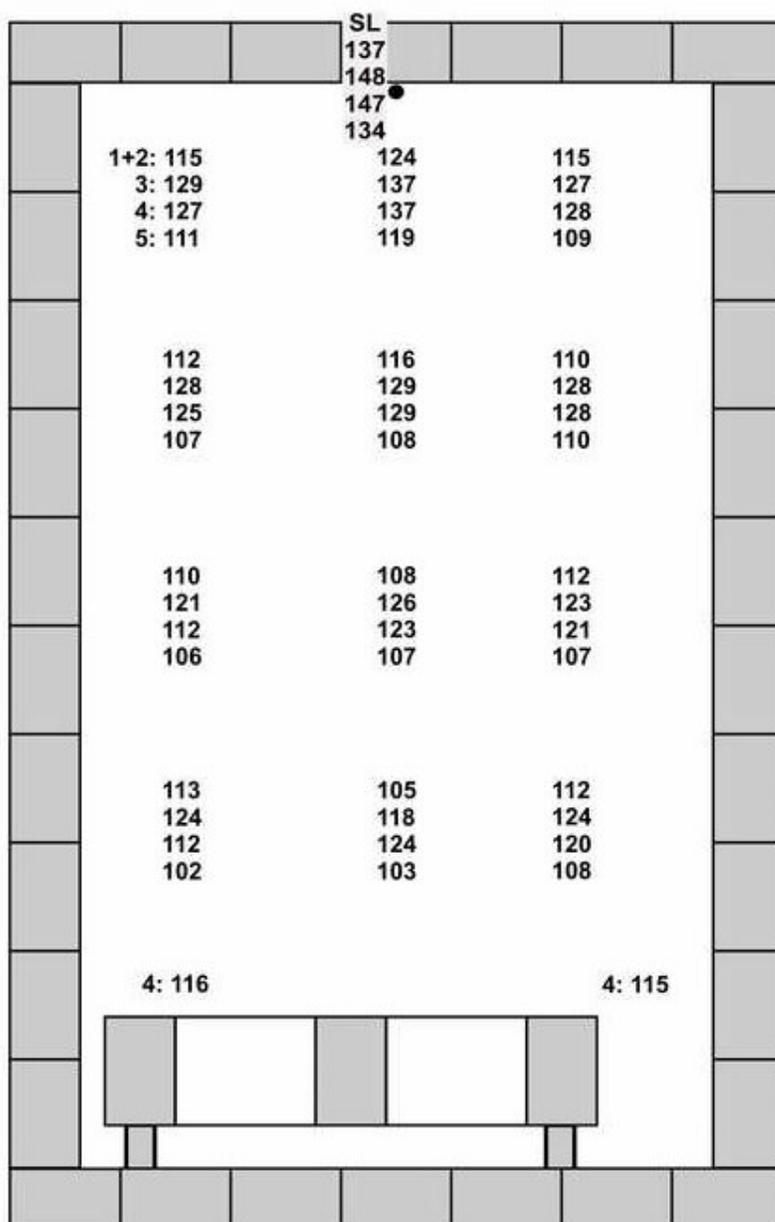


Figure 4

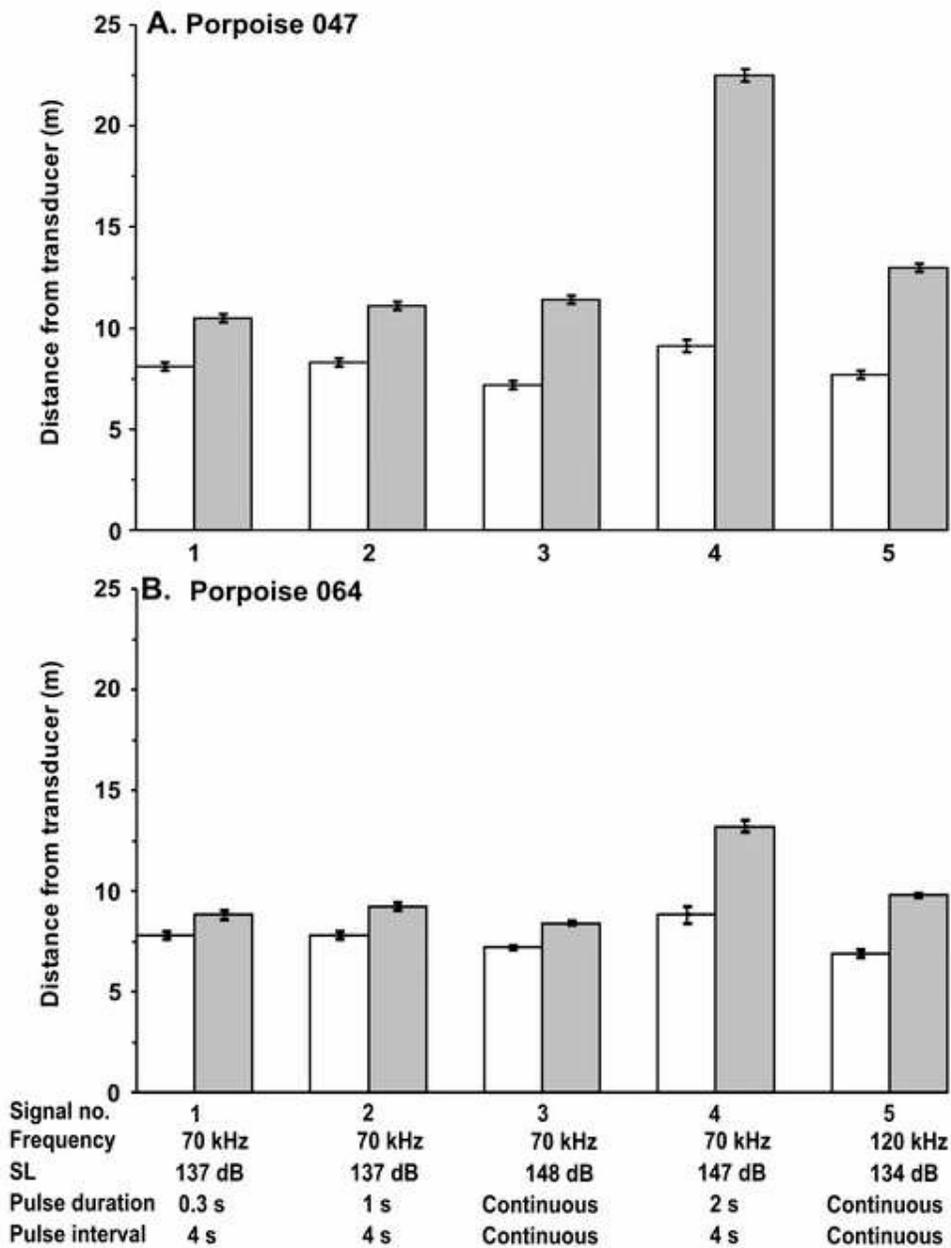
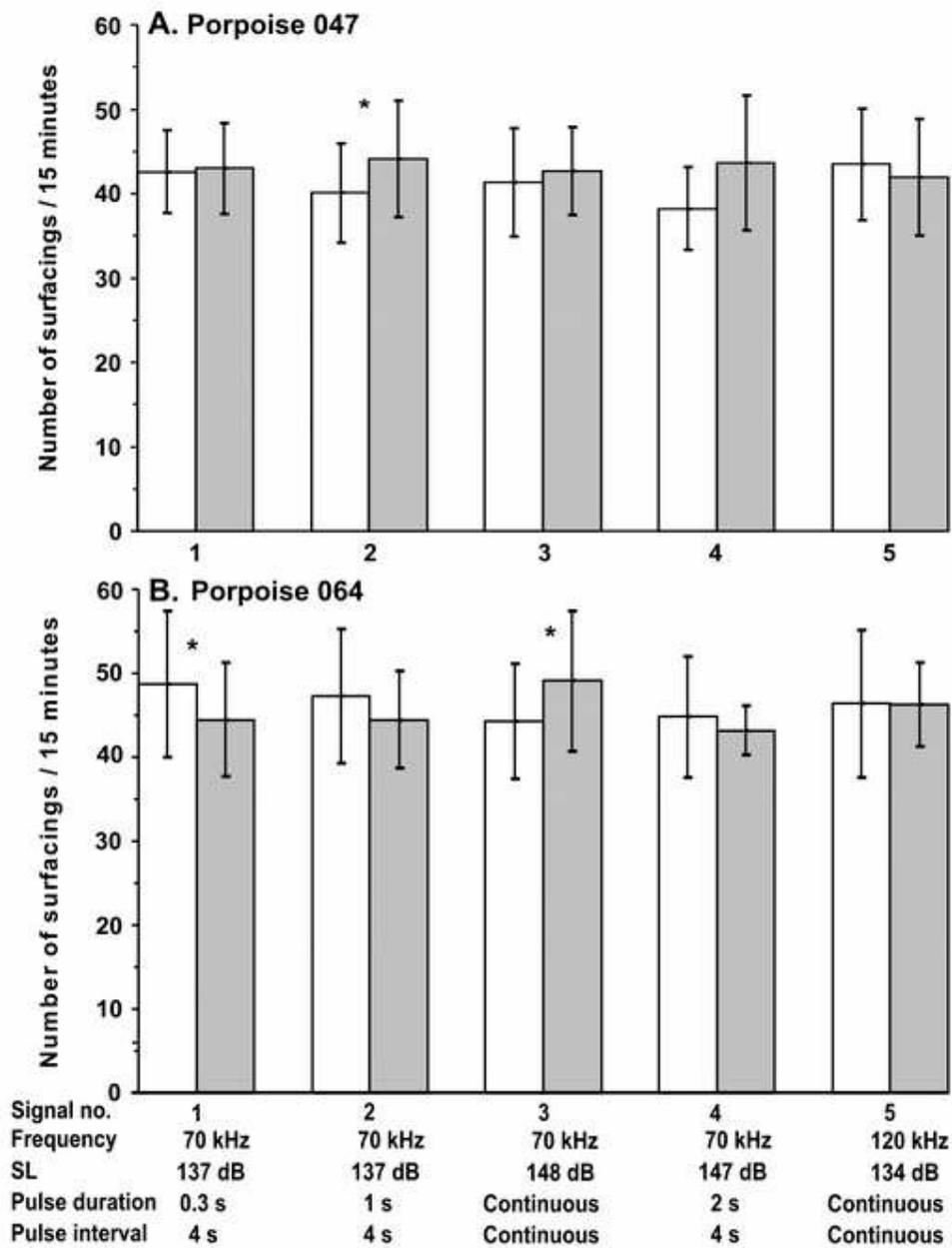


Figure 5



A