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Pelagic fish and harbour porpoise at North Sea wind farms: acoustic investigation and science communication

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CHAPTER 4



An experimental sound exposure study at sea: no spatial deterrence of free-ranging pelagic fish

AN EXPERIMENTAL SOUND EXPOSURE STUDY AT SEA: NO SPATIAL
DETERRENCE OF FREE-RANGING PELAGIC FISH

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Abstract

Acoustic deterrent devices are used to guide aquatic animals from danger or toward migration paths. At sea, moderate sounds can potentially be used to deter fish to prevent injury or death due to acoustic overexposure. In sound exposure studies, acoustic features can be compared to improve deterrence efficacy. In this study, we played 200–1600Hz pulse trains from a drifting vessel and investigated changes in pelagic fish abundance and behavior by utilizing echosounders and hydrophones mounted to a transect of bottom-moored frames. We monitored fish presence and tracked individual fish. This revealed no changes in fish abundance or behavior, including swimming speed and direction of individuals, in response to the sound exposure. We did find significant changes in swimming depth of individually tracked fish, but this could not be linked to the sound exposures. Overall, the results clearly show that pelagic fish did not flee from the current sound exposures, and we found no clear changes in behavior due to the sound exposure. We cannot rule out that different sounds at higher levels elicit a deterrence response; however, it may be that pelagic fish are just more likely to respond to sound with (short-lasting) changes in school formation.

Introduction

Human presence on earth has a detrimental impact on marine and freshwater environment, through exploitation and alteration of waterways for transport, fisheries, agriculture, industry, and recreation (Halpern et al., 2008; Österblom et al., 2017; Su et al., 2021). Several human activities may pose an unwanted physical challenge and even lifethreatening danger to aquatic animals (Panagiotopoulos et al., 2024). Examples of such hazardous activities are taking in water at pumping stations and hydroelectric power plants, and making loud sounds during seismic surveys, pile driving, or detonation of explosives (Putland and Mensinger, 2019; Slabbekoorn et al., 2010). Fish may suffer injury or death, due to impingement with screens and gates, entrainment in turbines and propellers, rapid water pressure changes, and acoustic overexposure with extreme sound levels (Andersson et al., 2016; Brown et al., 2014; Halvorsen et al., 2012; Piper et al., 2019). Placement of physical barriers is often not desirable or possible, and behavioral deterrence is therefore typically a preferred mitigation method, which may be possible through targeting their sensory system (Putland and Mensinger, 2019).

Sound may be a suitable modality to deter fish away from potentially dangerous locations. Sound propagates well in water and—depending on the source—omnidirectionally, unlike chemical cues, and its propagation is not affected by low light conditions or low visibility (Slabbekoorn et al., 2010). Several studies have reported on fish deterrence efficacy through acoustic deterrent devices (ADDs), with a reduction in mortality sometimes of more than 80% (Maes et al., 2004; Ross et al., 1996; Sonny et al., 2006). A relatively recent review showed that the majority of acoustic deterrence studies report an efficacy of at least 50% in deterring fish (Putland and Mensinger, 2019). However, the experimental design, in terms of sample sizes, controls periods, and replicate sites, is often suboptimal in such studies. There may also be a publication bias towards positive results, while the efficacy of ADDs is likely to vary per location, application, target species, habitat, and the acoustic characteristics of the device (Carretta and Barlow, 2011; Demuyne et al., 2024).

The efficacy of ADDs will depend on the match between sound stimuli used as well as on the hearing sensitivity of the target species (Putland and Mensinger, 2019) and the flight triggering potential. For example, in some fish species, predatory sounds have been shown to elicit a stronger fleeing response than pure tones across the same frequency range (Liu et al., 2019). In other prey species, sounds associated with predation may elicit a freezing or hiding response (Seigel et al., 2022). Artificial sounds with different temporal patterns have also been shown to elicit different response patterns. Sound treatments with noncontinuous sounds resulted in stronger behavioral responses and higher stress levels in various fish species (Neo et al., 2014; Vetter et al., 2015; Wysocki et al., 2006). Such data suggest that it may be useful to explore naturally meaningful sounds, but also artificially intermittent or pulsed sound stimuli to optimize ADDs (Hubert et al., 2020; Neo et al., 2014).

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Several studies have shown that experimental exposures with man-made sounds can affect the swimming depth of fishes (Hawkins et al., 2014; Kok et al., 2021; Neo et al., 2016), but horizontal avoidance or attraction effects are scarce (van der Knaap et al., 2021; van der Knaap et al., 2022). Most of the deterrence studies reviewed by Putland and Mensinger (2019) found changes in fish presence or deterrence locally or at a small spatial scale, and are thus less relevant for applications at sea. In contrast, large-scale fleeing is commonly found in marine mammals (Anderwald et al., 2013; Cure et al., 2016). ADDs can therefore successfully prevent harm to them, from hazardous human activities, or prevent predation of fish by them in offshore aquaculture or fishing nets (Brandt et al., 2013; Götz and Janik, 2013; Mikkelsen et al., 2017). For fish, the lack of horizontal spatial effects may often be explained by test conditions in enclosures. In situ experiments, using free-ranging fish with a large home range may have most potential to find deterrence effects, although it is challenging to collect observational data on response patterns at sea.

Echosounders are widely used in fisheries and are used to monitor the abundance and behavior of free-ranging fish in situ (Simmonds and MacLennan, 2008). Echosounders produce high-frequency sound, inaudible to fish, and monitor the echoes that are reflected from fish schools and individual fish. They are therefore often used in pelagic fisheries to guide trawling efforts, with the echosounder mounted in the hull of the ship (Benoit-Bird and Lawson, 2016; Warren, 2012). Echosounders have already been successfully used to examine the effects of anthropogenic disturbances on schooling fish at sea and in freshwater systems (Hawkins et al., 2014; Kok et al., 2021; Sonny et al., 2006). Long-term deployment of bottom-moored echosounders enable collection of longitudinal data of free-ranging fish—before, during, and after acoustic events. However, pelagic fish abundance has been shown to vary strongly over time and space. Therefore, sufficient baseline data, experimental exposure, spatial replication, and controls, are needed to investigate the impact of acoustic disturbances (Kok et al., 2021).

Pelagic fish are important to the ecosystem (Cury et al., 2000; Palomera et al., 2007; Stephenson and Smedbol, 2019), have large commercial value (Stephenson and Smedbol, 2019; Toresen and Østvedt, 2008), and may suffer from injury when close to seismic surveys, pile driving, or detonation of explosives (Casper et al., 2013; Popper and Hawkins, 2019; Slabbekoorn et al., 2019). There is some evidence that pelagic fish move away from the seismic survey sounds themselves and that fish can change their depth distribution during the survey (Engås et al., 1996; Jorgenson and Gyselman, 2009; Slotte et al., 2004). Tagging studies on benthic Atlantic cod (*Gadus morhua*) have also revealed delayed spatial responsiveness to an experimental seismic survey and subtle displacements in association with nearby pile driving (van der Knaap et al., 2021; van der Knaap et al., 2022), but tagging is less suitable for pelagic fish, as they may not stay long enough in an area to yield sufficient data. Whilst tag data provide information on individual behavior, echosounders yield insight into changes in abundance and behavior of fish in the water column at a specific locality (Hawkins et al., 2014; Kok et al., 2021; Sonny et al., 2006).

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In the current study, we examined the behavioral response of free-ranging pelagic fish to experimental sound playbacks at sea, aiming to deter fish. We monitored pelagic fish using a transect of bottom-moored echosounders and experimentally exposed the fish to sound treatments played back from a drifting vessel. We aimed to answer the following research questions: (1) Do experimental sound exposures at sea affect the presence and depth of pelagic fish? (2) Is it possible to distinguish tracks of individual fish from other floating objects or animals? (3) If so, do experimental sound exposures at sea affect the presence, swimming direction, speed, and depth of individual pelagic fish? We used two different sound pulse types for the experimental exposure and two different temporal patterns, to explore deterrence efficiency of stimuli and potential improvements.

Materials and methods

1. Location

We deployed a transect of four multi-sensor frames in the offshore windfarm C-Power, 30km off the Belgian coast (Figure 1). C-Power was finalized in 2013 and consists of 54 wind turbines with a total capacity of 325.2MW. It is part of a large zone of windfarms along both sides of the Belgian–Dutch border. The multi-sensor frames were deployed on January 14, 2022, and retrieved on March 18 and March 24, 2022. The frames were deployed on the sea floor at a depth of 19.5–25.5m and the distance between adjacent frames was 680–756m. We conducted acoustic exposure experiments from January 23 up to January 26, 2022. In this time of the year and at this location, we expected to detect the following species in the water column: European sprat (*Sprattus sprattus*), Atlantic herring (*Clupea harengus*), and whiting (*Merlangius merlangus*) (ICES, 2023; Van Ginderdeuren et al., 2014).

2. Multi-sensor frames

We used four bottom-moored, multi-sensor frames (1.6 x 1.2 x 1 m³; L x W x H) equipped with an echosounder, a hydrophone, and an acoustic release system (VR2AR, VEMCO, Billings, MT). The acoustic release system ensured a full recovery of the entire frame. Each stand-alone autonomous echosounder [wide band autonomous transceiver (WBAT), Kongsberg Maritime AS, Bergen, Norway] used an upward pointing wideband split-beam 70kHz (ES7018CD, Simrad, Horten, Norway) and a split-beam 200kHz (ES200-7CDK-split, Simrad) transducer. The echosounders were programmed to be active continuously from 08:00 until 19:30 (local time, UTC+1) and used the 70 and 200kHz transducers alternately in 15min blocks. The echosounders were calibrated using standard spheres (ICES, 2015). Depending on the frame, the hydrophone was either a SoundTrap (Ocean Instruments, Auckland, New Zealand), ST300, ST600 HF (high frequencies), or a ST4300 HF fourchannel recorder with four external HTI-96-MIN hydrophones (High Tech Inc., Long Beach, MS). The hydrophones were set to record sound at a sampling rate of 48kHz for either 10min/hr or continuously.

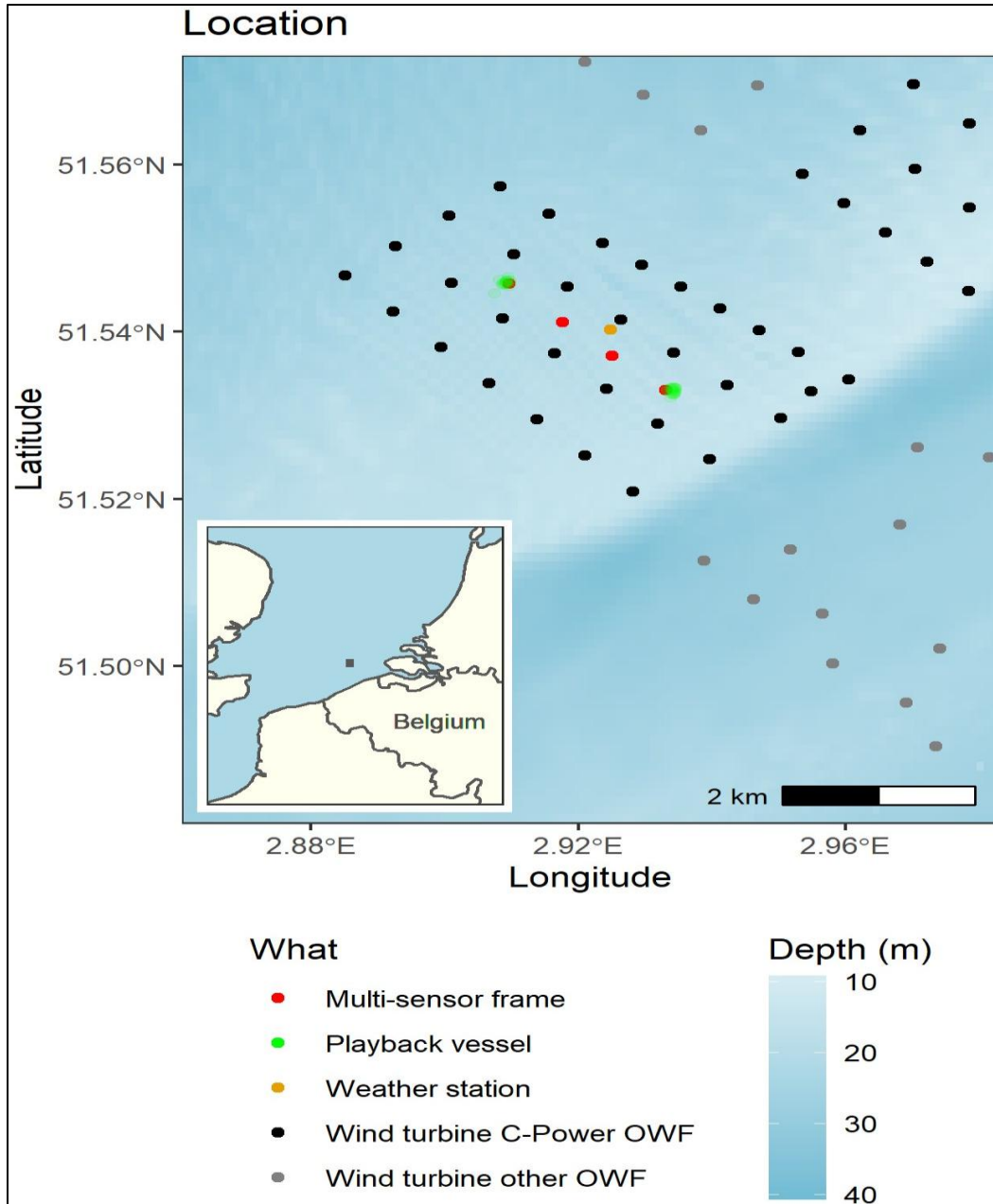


Figure 1. The location of the exposure experiment in the offshore windfarm (OWF) C-Power, 30km off the Belgian coast. The four multi-sensor frames, with echosounders and hydrophones, are indicated as red points and were placed on a linear transect, with distances between adjacent frames from 680 to 756m. The position of the vessel with the speaker during the exposure events is indicated as green points and alternated between the two sides of the transect. The distance between the vessel and the closest frame was on average 73m (σ 37m).

3. Experimental design

We made four day trips with the vessel Ephyra to play back a set of sound stimuli for experimental exposures of the locally present pelagic fish community. The playback alternated between two locations: just northwest of the northernmost frame and southeast

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of the southernmost frame. From at least 5min before the start of a trial until at least 5min after a trial, the playback vessel drifted with minimum engine power at on average 73m (σ 37) from the closest frame. Each day, we conducted eight sound treatment trials and five to seven control treatment trials during which we drifted without sound exposure. Most of the time (62% of the trials), we conducted one sound trial and one control trial at one playback location, and then moved to the other location to increase independence of the trials. It took 20min to transit between both playback locations at a speed of 4–5 kn. Part of the time (38% of the trials), we had to speed up because of upcoming bad weather, by performing two sound treatment trials and one control treatment trial per playback position. In total, we conducted 32 sound treatment trials and 26 control treatment trials. In each of the sound treatment trials, we played back one of four sound treatments. The order of the sound treatments was counterbalanced.

4. Sound treatments

Each sound treatment trial lasted 10min and consisted of 20 pulse trains of 10.2s. Each pulse train consisted of 36 pulses of 0.02s. The pulses (200–1600Hz) were either all linear up-sweeps or all white noise bursts [Figures 2(c) and 2(d)], the intervals between the pulses were either regular (interval always 0.27s) or first linearly decelerated and then accelerated (interval ranged from 0.05 to 0.50s) [Figures 2(a) and 2(b)]. This yielded four different pulse trains (2 pulse types 2 pulse interval types) and each sound exposure trial consisted of 20 pulse trains of the same type. The 20 pulse trains were separated with silence periods of 11.8–29.8s and the order of the silence periods was determined randomly during each trial. During the control trials, we just played back silence. The use of up-sweeps and silence periods was based on the FaunaGuard Fish ADD (Ace Aquatec, Dundee, UK). All treatments were generated using Audacity (version 3.0.5; Audacity Team, 2023). The treatments were played through a speaker (LL-1424HP, Lubell Labs, Whitehall, OH) at a depth of 8m below a surface buoy and 10m behind the drifting vessel. The speaker is omnidirectional in frequency range of the used pulse trains. The sound treatments were played back from a laptop, which was connected to the speaker via an amplifier (DIGIT 3K6, SynQ, GrootBijgaarden, Belgium) and transformer (AC1424HP, Lubell Labs). The selected level of the amplifier was based on the maximum level that yielded a clean signal without clipping by the speaker during a test in a river prior to the experiment. At sea, the sound pressure levels (SPLs) were determined using the hydrophones at the multi-sensor frames and depended on the distance from the playback vessel. The root-mean-square (rms) SPL (100–2000Hz) varied mostly between 112 and 133dB re 1 μ Pa depending on the distance to the source, with ambient levels of between 106 and 112dB re 1 μ Pa during silence playbacks, calculated over 10.2s periods [Figure 2(f)].

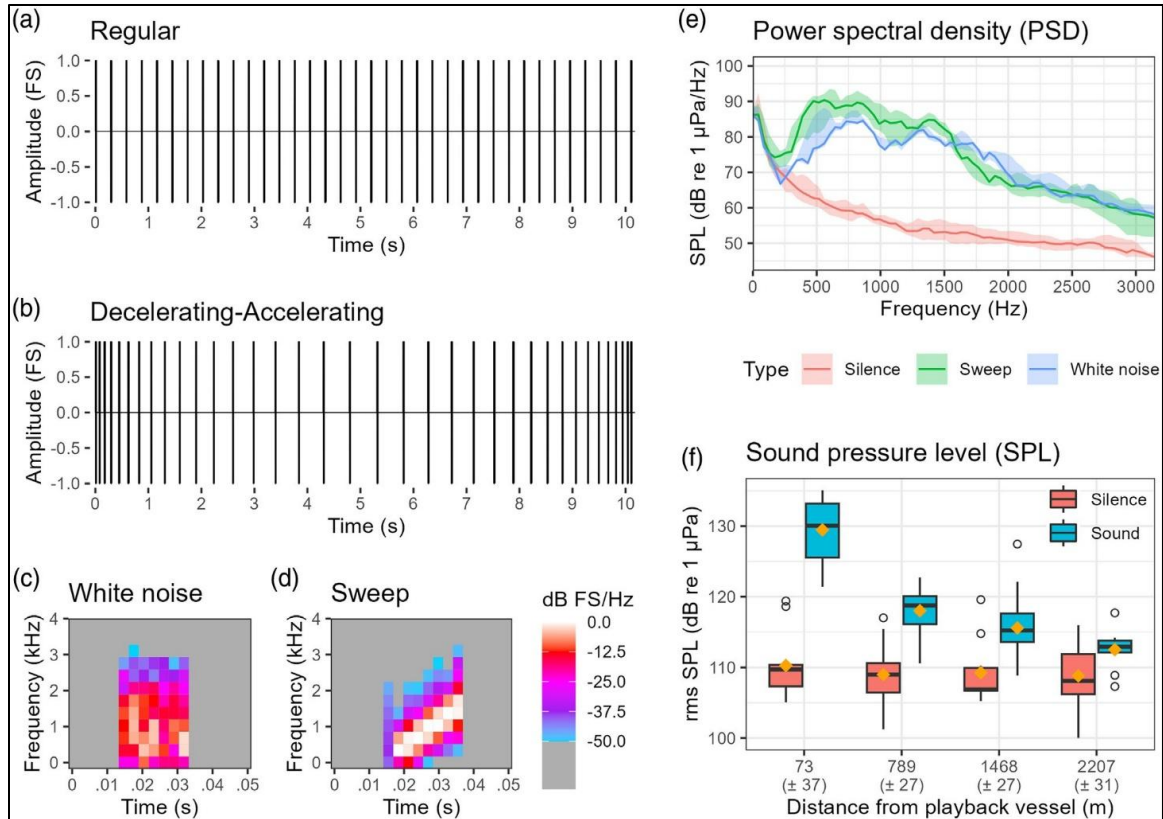


Figure 2. Overview of the sound treatments and recorded spectra and levels. We played back 10.22s (a) regular and (b) decelerating–accelerating spaced pulse trains (waveforms). Each pulse train consisted of 36 pulses of 0.02s, either (c) white noise bursts or (d) sweeps, resulting in four different sound treatments. The pulses were generated in the 200–1600Hz frequency range (spectrograms, window type: Hann; window length: 128; overlap: 0.02%). (e) The resulting power spectral density plot shows the spectra of the recorded sound and control conditions at 73m (σ 37) from the drifting playback vessel (window type: Hann; window length: 1024; overlap: 50%). (f) The sound pressure levels (100–2000 Hz bandpassed) during the control silence playbacks (red) were similar at all frames and therefore represented the ambient level. The levels of the recorded sound playback (blue) decreased with increased distance from the playback vessel, but remained above mean ambient background noise at all frames. Each distance on the x axis represents one of the frames, and the yellow diamonds indicate the means.

5. Data processing and statistics

We processed the raw echosounder data using LSSS (Large Scale Survey System; version 2.14, MAREC, Bergen, Norway) to ultimately gain insight into fish abundance and behavior during days with sound exposures. The procedure employed to process the echosounder data were identical for both echosounder frequencies. We used automatic functions of LSSS to detect the surface line, fish marks, as well as individual tracks of single targets (fish or other objects). All echograms were manually validated for the surface line and for the false positives and negatives on fish marks. Fish marks consisted of marks associated with individual fish and small to, occasionally, medium aggregations. Next, we exported the scored fish marks, the area backscattering coefficient (s_a ; Maclennan et al., 2002), integrated

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for each 0.5m of the water column per 10s, and acoustic target detections for all tracked objects as x, y, and z positions and associated target strength (TS) (Maclennan et al., 2002).

The data were further processed in R (version 4.3.0; R Core Team, 2023). To determine the effects of the sound treatments, we used 5min bins before, during, and after the sound exposures or silent controls. Each full exposure period lasted 10min, so we used two 5min during-bins (labelled during 1 and 2). For these periods and for each individual echosounder, we determined whether fish were present based on the scored fish marks, the sum of the area backscattering coefficient as a proxy for biomass, and the depth center of (bio)mass (Kok et al., 2021). For the individual tracks, we found that the majority of the tracks followed the tidally dependent pattern of current speed and direction. Hence, we used the direction of the tracks, and tidal current direction from a nearby weather station (Figure 1; MFC OD Nature RBINS, 2022) to determine the orientation of the bottom-moored frames. We used the target strength of the tracks to distinguish between fish (60dB FS) and other objects (<60dB FS)—a procedure that is further explained in the results. For all time bins (before, during 1 and 2, after), we determined whether at least one fish track was detected, their circular mean swimming direction, mean speed, and mean depth. For each period, we also determined the proportion of time that the echosounder used the 70kHz transducer, to account for differences between the two transducers.

We analyzed differences in fish presence and behavior between periods to determine the effect of the sound exposures. We used generalized linear models (GLMs) with a binomial distribution and logit link-function for the presence/absence data, Watson–Wheeler tests for the swimming direction data, and analysis of variances (ANOVAs) for all other response variables. We used the period (before, during 1 and 2, after) and the “proportion 70 kHz” as covariates in the full model. We made separate models for sound treatment and control treatment trials. Based on the AICc (Akaike Information Criterion with a correction for finite sample sizes) score, we determined whether “proportion 70 kHz” contributed to the model fit and was included in the final model. To determine the effect and significance of the covariates, we ran the final models. When we found a significant effect of period, we used a Tukey’s post hoc test to make pairwise comparisons between the periods. All analyses and plotting was done using R-packages MuMIn (Barton, 2016), and multcomp (Hothorn et al., 2008), circular (Agostinelli and Lund, 2023), and ggplot2 (Wickham, 2016).

6. Ethical notes

We observed wild free-ranging animals in situ and exposed them to sound at levels that could not cause physical harm.

Results

We conducted 32 sound treatment trials and 26 control treatment trials during which fish were monitored using four multi-sensor frames. However, we only detected fish in a minority (33.3%) of the treatment periods (before, during 1 and 2, and after) of all trials [Figure 3(a)]. The number of 5min bins with at least one fish mark was not affected by treatment period, neither in control trials [$\chi^2 = -1.975$; $df = 412, 415$; $p\text{-value} = 0.578$; Figure 3(a)] nor in sound exposure trials [$\chi^2 = -2.788$; $df = 507, 510$; $p\text{-value} = 0.426$; Figure 3(a)]. The area backscattering coefficient, a proxy for biomass, was also not affected, either by combining all frames [control trials: $F_{(3,124)} = 0.980$; $p\text{-value} = 0.405$; sound exposure trials: $F_{(3,160)} = 0.968$; $p\text{-value} = 0.409$; Figure 3(b)], or by only considering the frames closest to the speaker [control trials: $F_{(3,28)} = 0.259$; $p\text{-value} = 0.854$; sound exposure trials: $F_{(3,31)} = 0.370$; $p\text{-value} = 0.775$; Figure 3(c)]. The depth of fish biomass was also not affected by treatment period, a result consistent for all frames together [control trials: $F_{(3,124)} = 0.250$; $p\text{-value} = 0.861$; sound exposure trials: $F_{(3,159)} = 0.990$; $p\text{-value} = 0.399$; Figure 3(d)] or just for the frame closest to the speaker [control trials: $F_{(3,28)} = 0.210$; $p\text{-value} = 0.889$; sound exposure trials: $F_{(3,31)} = 0.123$; $p\text{-value} = 0.946$; Figure 3(e)].

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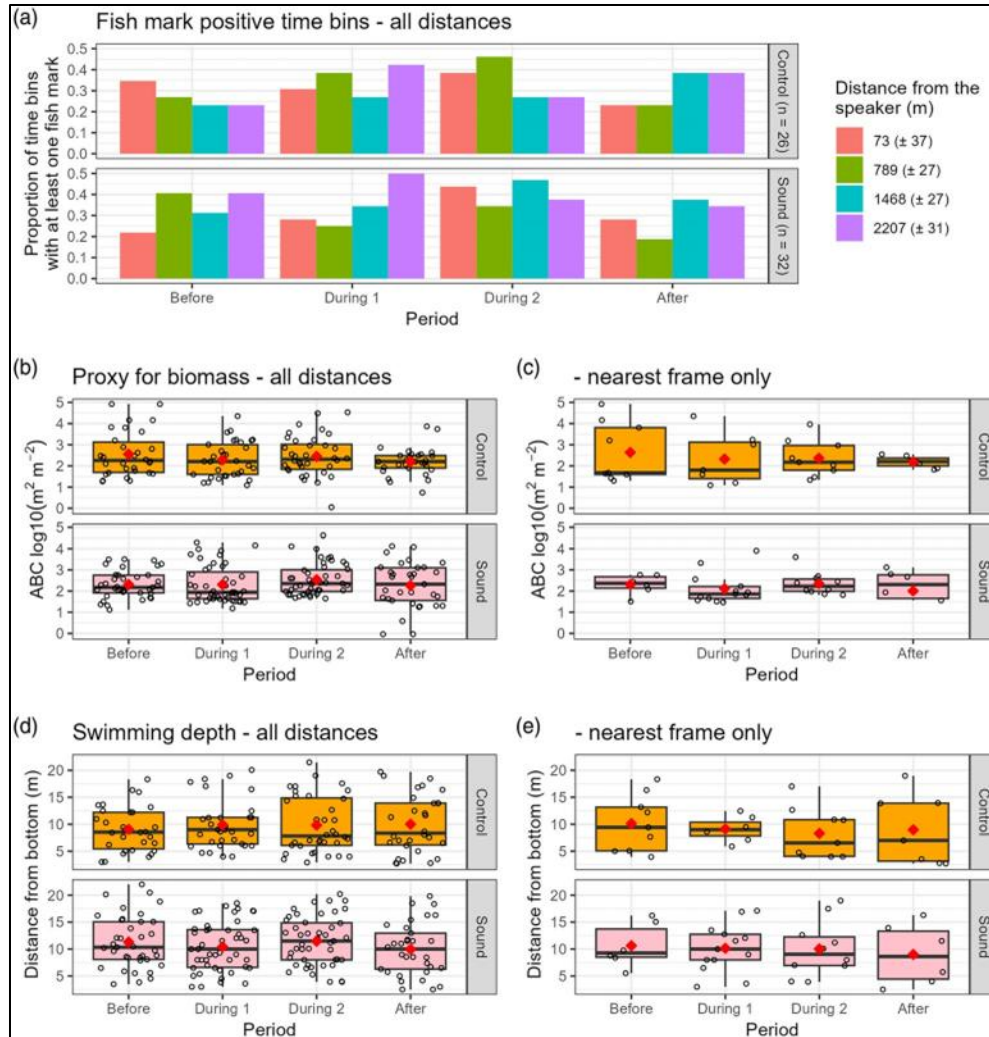


Figure 3. Fish marks 5min before, during (two 5min bins), and after the experimental exposure events. (a) Proportion of trials in which fish presence was detected, indicated for all echosounder distances from the playback vessel (color). (b) Sum of area backscattering coefficient (a proxy for biomass) per trial and echosounder. (c) Similar to plot b, but only with the data of the echosounder closest to the speaker (\bar{x} 73m, σ 37m). (d) Mean depth center of mass (distance from the bottom) per trial and echosounder. (e) Similar to plot d, but only with the data of the echosounder closest to the speaker (\bar{x} 73m, σ 37m). The box-whisker plots indicate the median, first, and third quartile, and min and max excluding outliers. The red diamond symbols indicate the means of each distribution.

The targets that were tracked followed a tidal pattern in terms of speed and direction [Figures 4(a) and 4(b)]. The TS of the tracks followed a bimodal distribution with a cutoff at -60dB FS [Figure 4(c)], suggesting two distinct types of tracked targets. Moreover, tracks with a lower TS were associated with a lower deviation from current speed [Figure 4(d)] and direction [Figure 4(e)]. These targets were hence not classified as fish and could be seaweed, abiotic objects, or planktonic animals, like jellyfish. Objects with a higher TS were classified as fish. Most tracks classified as fish were detected in the at dawn, and, to a lesser

extent, at dusk [Figure S1(b)]. Consequently, a lesser amount of fish biomass was detected at the time of our trials during the day (~09:40–16:40 UTC +1).

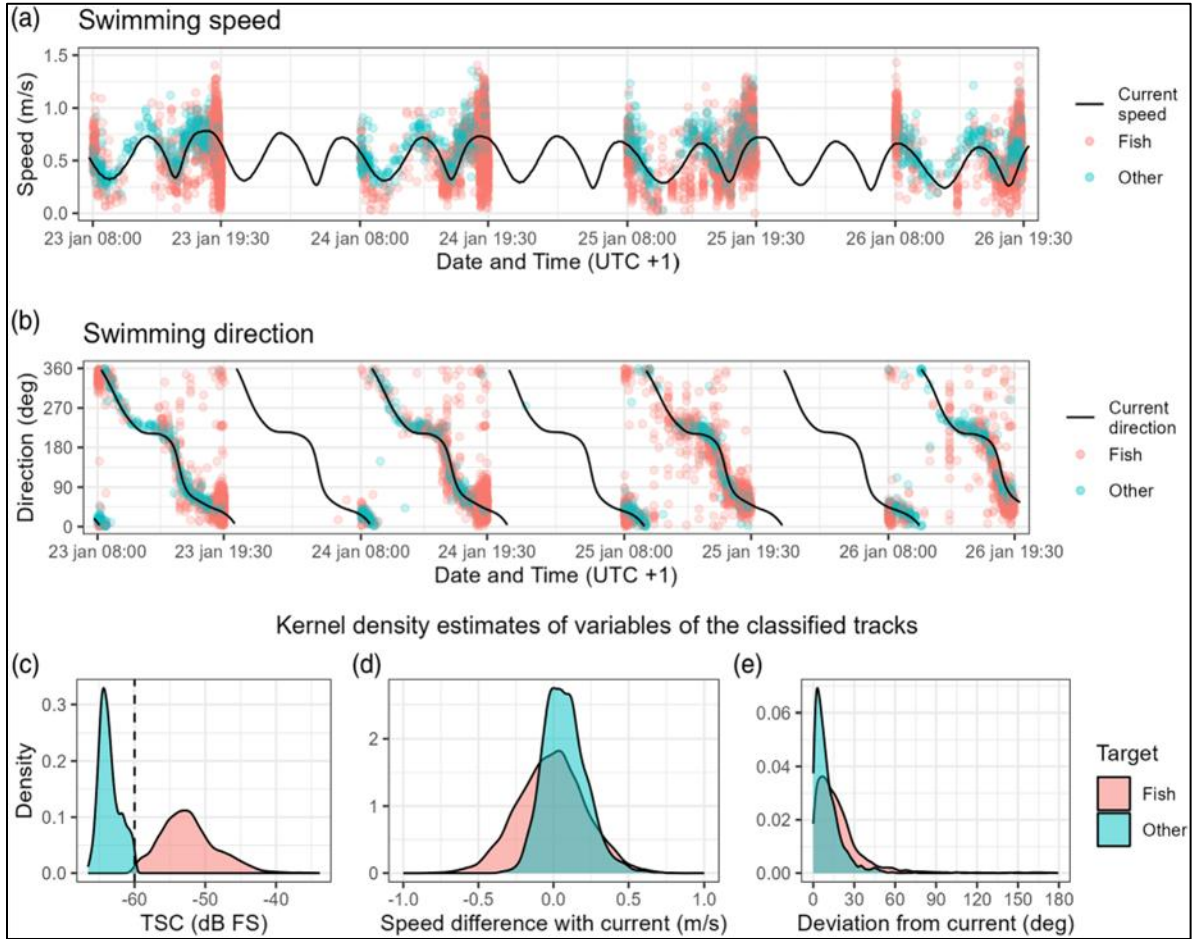


Figure 4. (a) The swimming speed (b) and direction (b) of all detected tracks over time, the color indicates whether a track was classified as fish or another target. We plotted the 4 days during which we conducted experimental sound exposures. The echosounders were active from 8:00 to 19:30 (UTC +1; local time) and all trials were performed between 09:40–16:40 (UTC +1). The solid black line indicates (a) the water current speed and (b) direction. (c)–(e) Below, the kernel density estimates show the distributions of metrics for the tracks that were classified as fish and other targets. (c) This includes the mean beam compensated target strength (dB FS), (d) the difference of the targets' swimming speed from the current speed, (e) the difference of the targets' direction from the current direction.

Fish tracks were detected in 19.6% of the number of 5min bins and this was not affected by treatment period, neither during control trials [$\chi^2 = -1.677$; $df = 395398$; p -value = 0.642; Figure 5(a)] nor sound exposure trials [$\chi^2 = -2.0235$; $df = 492495$; p -value = 0.568; Figure 5(a)]. The swimming direction of the fish tracks did not change significantly over the treatment periods [control trials: $W = 2.847$; p -value = 0.828; sound exposure trials: $W = 4.701$; p -value = 0.583; Figure 5(b)]. The swimming speed of the individual fish was also not

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affected by treatment period, when considering the data from all frames [control trials: $F_{(3,79)} = 0.112$; p-value = 0.953; sound exposure trials: $F_{(3,83)} = 1.186$; p-value = 0.320; Figure 5(c)], nor when considering data from the frames closest to the speaker [control trials: $F_{(3,19)} = 0.068$; p-value = 0.976; sound exposure trials: $F_{(3,25)} = 0.095$; p-value = 0.962; Figure 5(d)]. Notably, the swimming depth of fish did change during control trials [$F_{(3,80)} = 5.607$; p-value = 0.002; Figure 5(e)] and pairwise comparisons showed that fish detected after the before period swam significantly higher in the water column [p-values ≤ 0.0332 ; Figure 5(e)]. During sound exposure trials, we also found a change in swimming depth [$F_{(3,88)} = 6.033$; p-value = 0.001; Figure 5(e)]. Here, the swimming depth was significantly lower in the after period compared to the before and first 5min of sound exposure [p-values ≤ 0.003 ; Figure 5(e)]. A non-significant trend was found for the decrease in depth between the second half of the exposure and the after period. At close range, there was also significant variation in swimming depth during the control trials [$F_{(3,19)} = 4.099$; p-value = 0.021; Figure 5(f)]: the swimming depth before the silent control exposure was significantly lower than in the after period (p-value = 0.019; Figure 5(f)), and this was a trend when comparing the before period with during 2 [p-value = 0.0654; Figure 5(f)]. However, for the sound exposure trials at close range, no significant changes in swimming depth between periods were observed [$F_{(3,25)} = 0.803$; p-value = 0.504; Figure 5(f)].

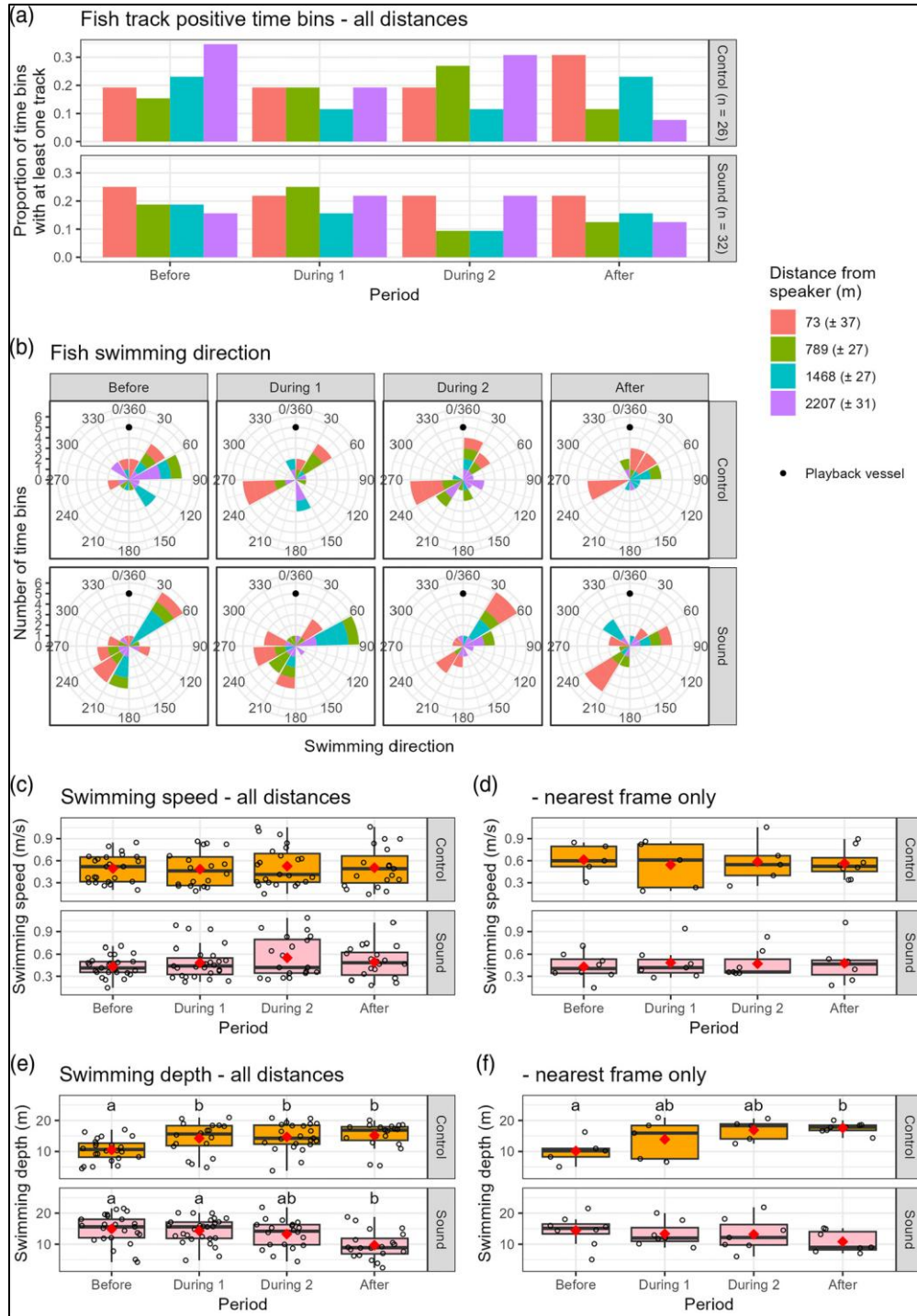


Figure 5. Fish tracks 5min before, during (two 5min bins), and after the experimental playbacks. (a) Proportion of trials in which at least one fish track was detected, indicated for all echosounder distances from the playback vessel (color). (b) Direction of the fish tracks relative to the direction to the playback vessel (0 degrees angle). The circular stacked bars are mean swimming directions per trial period for each echosounder (color). (c) The mean swimming speed per trial for all echosounders, and (d) only for the nearest. (e) The mean swimming depth (distance from the bottom) per trial for all echosounders, and (f) only for the nearest. Significant differences between periods are indicated by different letters above boxplots.

Discussion

We performed an experimental exposure study with sound playbacks of artificial pulse trains from a drifting vessel at sea and monitored the presence and behavior of freeranging pelagic fish. The goal was to explore and improve deterrence efficiency of stimuli for ADDs. However, the number of fish marks, their biomass, and depth were not affected by the exposures. Tracks from individual fish also did not reveal any avoidance or changes in swimming speed, but we found some changes in swimming depth. We were unable to test for significant variation in efficacy among stimulus variants, as acoustic responsiveness was low or absent and during a considerable number of trials, no fish were detected.

1. Lack of spatial response to sound

We did not find evidence that fish were laterally deterred by the current sound exposures in any way. The number of 5min bins with fish marks, the fish biomass, the number of 5 min bins with individual fish tracks, and the direction of individual fish tracks—all remained similar from before to during and after the sound exposures. Some studies have reported spatial effects of sound on free-ranging fish in a river context, to guide migration (Perry et al., 2014; Sand et al., 2000), or to keep fish away from a water inlet (Maes et al., 2004; Sonny et al., 2006). However, studies in more open water are limited to largescale reports on density changes or fishing impact related to seismic survey sounds (Engås et al., 1996; Jorgenson and Gyselman, 2009; Slabbekoorn et al., 2019; Slotte et al., 2004). Pelagic fish in schools may respond to extreme or unfamiliar sound events as in other threatful situations. Pelagic schools typically respond to predators by changing school formation and making rapid movements, but not necessarily moving away from the predator (Gerlotto et al., 2006; Pertzalan et al., 2023). We did not detect any changes that indicate changes in fish marks, but some changes may be too short-lasting and limited to the sound onset to be detected in the current setup (Neo et al., 2014, 2016; Shafiei Sabet et al., 2016).

Fishing vessels may be the most common threat to fish, which also make sound, and thereby potentially affect (spatial) behavior of fishes (De Robertis and Handegard, 2013; Kaartvedt et al., 2012). Bottom trawling vessels have been reported to induce a down-diving response for cod (*Gadus morhua*), depending on the water layer, also followed by a lateral movement away from the sound (Handegard et al., 2003). However, other studies report changes in schooling morphology and very little change in depth and lateral dispersal (Gerlotto et al., 2004). To gain a better understanding of fish movement patterns relative to vessels, it has been suggested that measuring sound pressure levels are likely insufficient, and that particle motion levels should be considered (De Robertis and Handegard, 2013). We conducted our experimental exposure experiment from a vessel, but movement and engine activity were kept at a minimum during the experiment. It is likely that, for our results, the sound pressure levels and particle motion levels correlate well over distance due to the water depth (19.5m), implying similar conditions to free-field for our sounds (>200Hz) (Jansen et al., 2019; Nedelec et al., 2016).

2. Variation in swimming depth

We found significant variation in swimming depth of individually tracked fishes, both during control trials and during sound exposure trials. In control trials, fish swam less deeply during and after the silence exposure period. In the sound exposure trials, fish swam deeper after the sound exposure. The latter may indicate a relatively late response to the sound exposure, or to the end of the exposure, but we are cautious with drawing this conclusion as we also found significant variation in swimming depth in the control trials without sound exposure. Large variation in swimming depth has been reported before and highlights the need for good replication (Kok et al., 2021). Nevertheless, a diving response to sound is commonly reported in captive fish, and occasionally free-ranging fishes (Doksæter et al., 2012; Fewtrell and McCauley, 2012; Hawkins et al., 2014; Hubert et al., 2020; Kok et al., 2021; Peña and Ratalal, 2019; Sara et al., 2007), including return to pre-exposure baseline levels during the exposure (Neo et al., 2014). Fishes have also been reported to increase their depth in response to predator cues (Luca and Gerlai, 2012), novel environments (Cachat et al., 2010), weather conditions (Kaartvedt et al., 2017; Kok et al., 2021), and chemical pollution (Israeli-Weinstein and Kimmel, 1998). Altogether, the diving response seems to be a general fear or anxiety response (Jesuthasan, 2012), potentially as a mechanism to avoid danger.

3. Recommendations for future studies

We clearly showed that fish were not deterred from the exposure area and no substantial responses occurred in response to the sound exposures. We were unable to compare the efficacy of the different types of sound exposures in eliciting a behavioral response, because fish aggregations or individual fish were only detected in a minority of the time bins, both before and after the sound onsets. For future studies, we therefore advise to use longer periods before, during, and after the sound exposures, or to plan trials at the time and location of high fish abundance. Regarding our study, an alternative setup could have been to position the monitoring frames in a square. Such a setup would have allowed shorter transiting for replication across both sides of the study area but with sampling at smaller distances from the vessel. Several differences in response potential to acoustic stimuli have been found in controlled conditions (Hubert et al., 2022; Neo et al., 2014). Verifying these in situ and across species can benefit ADD development and efficacy.

ADDs have been applied to fish, especially in river contexts (Putland and Mensinger, 2019). The reports available seem promising, but we should be cautious for a potential publication bias. Furthermore, the number of studies is low and replications and experimental controls are often not optimal. The application at sea in marine mammals has proven to provide some success (Boisseau et al., 2021; Voß et al., 2023), even though habituation or conditioning may cause efficacy to fade (Carretta and Barlow, 2011; Neo et al., 2018). This may be less of an issue if the ADD sounds are linked to harmful consequences (Bejder et al., 2006). The application of ADDs for fish at sea and especially small pelagic species may have

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limited potential, as (pelagic) fish do not generally respond to threats by fleeing over far distances (Gerlotto et al., 2006; Pertzalan et al., 2023).

Conclusions

For the current study, we exposed free-ranging fish at sea to different sound treatments from a drifting vessel, and we examined fish abundance and behavior using a transect of bottom-moored frames with echosounders. The sound treatments were still detected by hydrophones at the furthest frames at 2.2km from the speaker. Nevertheless, we did not detect any changes in fish abundance nor behavior during the sound exposure trials. We found changes in swimming depth of individual fish, but also during the silent control trials. While we cannot rule out the possibility that different sound treatments at higher levels trigger a deterrence response, it is also possible that pelagic fish do not flee, but rather modify their schooling behavior momentarily in response to sound.

Supplementary material and data availability

For supplementary material of this article see the publication:
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All data used for the analyses reported in this article are available from the Zenodo repository, DOI:10.5281/zenodo.10598391.

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