Marine Pollution Bulletin 97 (2015) 111–117

Contents lists available at ScienceDirect

Marine Pollution Bulletin

journal homepage: www.elsevier.com/locate/marpolbul

Impulsive sounds change European seabass swimming patterns: Influence of pulse repetition interval

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ARTICLE INFO

Article history:
Received 16 April 2015
Revised 9 June 2015
Accepted 11 June 2015
Available online 15 June 2015

Keywords:
Dicentrarchus labrax
Sound temporal structure
Anthropogenic noise
Fish behaviour
Habituation

ABSTRACT

Seismic shootings and offshore pile-driving are regularly performed, emitting significant amounts of noise that may negatively affect fish behaviour. The pulse repetition interval (PRI) of these impulsive sounds may vary considerably and influence the behavioural impact and recovery. Here, we tested the effect of four PRIs (0.5–4.0 s) on European seabass swimming patterns in an outdoor basin. At the onset of the sound exposures, the fish swam faster and dived deeper in tighter shoals. PRI affected the immediate and delayed behavioural changes but not the recovery time. Our study highlights that (1) the behavioural changes of captive European seabass were consistent with previous indoor and outdoor studies; (2) PRI could influence behavioural impact differentially, which may have management implications; (3) some acoustic metrics, e.g. SEL_{cum}, may have limited predictive power to assess the strength of behavioural impacts of noise. Noise impact assessments need to consider the contribution of sound temporal structure.

1. Introduction

The ever increasing global energy demand has led to extensive exploitation of seas and oceans for both fossil and sustainable energy resources (EIA, 2013). Related human activities, such as seismic surveys and offshore constructions for wind farms and oil rigs, generate a substantial amount of noise in the underwater environment. This introduction of anthropogenic noise into the underwater acoustic scene may post a threat to aquatic life, including fish, causing a range of negative effects, from physical injuries in close range, to behavioural changes further away from the sound sources (Popper and Hastings, 2009a,b; Slabbekoorn et al., 2010). To ensure the stability of marine ecosystems under increased pressure of ocean exploitation, it is important to understand whether and how underwater anthropogenic noise may affect fish behaviour, which in turn may have consequences on fish populations.

Whether behavioural changes will result in negative fitness consequences, depends partly on whether fish habituate to the noise exposures and recover from the changes. However, behavioural observations in previous noise impact studies generally did not last long enough to show recovery after initial behavioural changes (Gerlotto and Fréon, 1992; Handegard et al., 2003; Doksæter et al., 2012; Fewtrell and McCauley, 2012; but see Neo et al., 2014). Moreover, a recovery or a decrease in response does not necessarily denote habituation, where the animals hear selectively while filtering out repeated or irrelevant sound signals in the background (Rankin et al., 2009). A decrease in behavioural response could also be attributed to (1) sensory adaptation, i.e. the sensitivity of the hearing organs is reduced by loud exposures, leading to temporary threshold shift (TTS), or (2) motor fatigue, i.e. animals become unresponsive due to exhaustion (Domjan, 2010). It is crucial to determine the mechanism of such behavioural recovery since the different mechanisms vary in their ecological implications.

Underwater noise impact assessments are complex also because anthropogenic noise shows a variety of amplitudinal, spectral and temporal variations. Of these, the temporal structure of sound is rarely studied, even though it may play a crucial role in triggering behavioural response in fish (Nelson and Johnson, 1972; Neo et al., 2014). For example, Neo et al., 2014 showed that European seabass (*Dicentrarchus labrax*) recovered more slowly from impulsive sounds than from continuous sounds (despite the former having lower accumulated sound pressure level), after exhibiting consistent initial behavioural changes upon noise exposures. Considering that impulsive sounds differ in various temporal features, there is a need for systematic studies addressing other temporal parameters, such as pulse repetition interval, pulse

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http://dx.doi.org/10.1016/j.marpolbul.2015.06.027
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repetition regularity, pulse duration and pulse shape (including rise time).

Among these temporal parameters, pulse repetition interval (PRI), which can also be expressed in pulse repetition rate (PRR, where PRR = 1/PRI), is rather variable among the current practices in pile driving and seismic surveys. PRI generally varies between 1–4 s (Matuschek and Betke, 2009) for pile driving and 5–15 s (McCauley et al., 2000) for seismic surveys. Different PRIs have been shown to influence the habituation rate to repeated sound stimuli in zebrafish and rats (Chanin et al., 2012; Davis, 1970). However, it is unclear if PRI also contributes to fish habituation to impulsive anthropogenic sound exposures, such as pile driving and seismic shootings.

In this study, we used a similar setup as in Neo et al., 2014 to answer two questions: (1) How do impulsive sounds of different PRIs (0.5 s, 1.0 s, 2.0 s, 4.0 s) affect the swimming patterns and behavioural recovery of European seabass? (2) Can the behavioural recovery be attributed to habituation? We expected larger PRIs to prolong the behavioural recovery and the recovery be attributed to habituation.

2. Materials and methods

2.1. Animal maintenance

The European seabass (mixed sex; 20–25 cm in total body length) came from a commercial hatchery (Ecloserie Marine, Gravelines, France) and were kept in four round holding tanks (diameter: 2.2 m; depth: 1 m) before and after the test trials at the Sea Mammal Research Company (SEAMARCO) in Wilhelminadorp, The Netherlands. Water was refreshed continuously with a recirculating system connected to the nearby Oosterschelde marine inlet and the water temperature varied from 4 to 12 °C throughout the experimental period (May–June 2013). Fish were fed pellets (Le Gouessant Aquaculture, Lamballe, France) every other day based on a temperature-dependent prescription. All experiments were performed in accordance with the Dutch Experiments on Animals Act and approved by the Animal Experiments Committee at Leiden University (DEC no: 13023).

2.2. Experimental arena

The experiment was conducted in a large outdoor rectangular basin (7 × 4 × 2 m) equipped with a water recirculating system at SEAMARCO. During the exposure trials, fish were put in a white nylon net enclosure (4 × 1.6 × 2 m) to ensure full coverage by an underwater video camera for observation (Fig. 1). A white tarp was placed at the bottom and in the background to ensure sufficient contrast in video images, without disrupting the normal swimming behaviour of the fish. Beside the basin, there was a research cabin containing sound generating and video monitoring equipment.

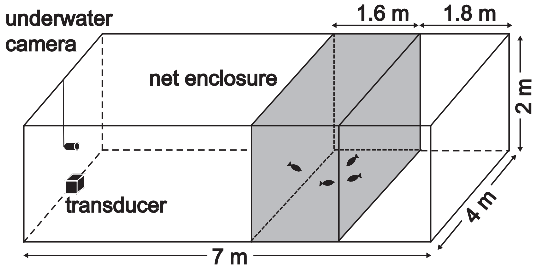

Fig. 1. Experimental basin at SEAMARCO. Shaded area is the net enclosure with restricted swimming space for four fish.

2.3. Treatment series

We exposed the fish to a series of four regularly repeated impulsive sound treatments differing in PRI: 0.5 s, 1.0 s, 2.0 s and 4.0 s (Table 1). The pulse duration of all the treatments was the same, which was around 0.15 s (Fig. 2a). The pulses were created in Adobe Audition 3.0 using filtered brown noise (band-passed: 200–1000 Hz; matching the hearing range of European seabass) and played back with an underwater transducer (LL-1424HP, Lubell Labs, Columbus, US) from a laptop through a power amplifier (Macro-tech 5000 VZ, Crown Audio, Elkhart, US). The whole experimental arena had a very homogenous sound pressure field during the playback of broadband sounds (Neo et al., 2014). The average root-mean-square sound pressure level (SPL_{rms}) before the exposure (ambient) in the experimental basin was 104 dB re 1 μPa, which was comparable to the ambient noise levels of our measurements in the Oosterschelde marine inlet. To quantify the amplitude level of the impulsive sound treatments, single-strike sound exposure level (SEL_{ss}) and zero-to-peak sound pressure level ($SPL_{z\text{-}p}$) were measured (Table 1). The amplitude levels were chosen to represent received level of pile driving at a range of around 50–100 km according to ideal cylindrical spreading. Spectral investigation confirmed that most of the sound energy of the pulses was concentrated between 200 and 1000 Hz (Fig. 2b).

Particle motion may be perceptually dominant in European seabass hearing (Popper and Fay, 2011), but we were unable to measure this. However, we believe that the lack of this information is not a concern in this study, since our aim was not to assess absolute threshold levels that can be extrapolated to outside conditions. Our main interest was to compare the effects of PRI on behavioural response while keeping other acoustic parameters constant.

2.4. Experimental set-up

We tested twelve groups of four fish, where each group was exposed to all four treatments ($N = 12$, 48 fish). The order of the treatments per fish group followed an incomplete counterbalanced design (12 of 24 possible orders), to minimise the potential 'carry-over' effect due to sequential exposures. At least 17 h prior to the trials, each fish group was transferred to the experimental basin to allow acclimatisation. 30 min before each trial, the transducer and the lights above the experimental basin were turned on. We conducted two trials per day: one in the morning and one in the afternoon, with a break of at least three hours in between. There was no external anthropogenic noise or disturbance near the study area during the trials. The trials consisted of 10 min of pre-exposure silence and 60 min of sound exposure. Based on pilot and previous studies (Neo et al., 2014), we expected the fish behaviour to recover within 60 min of sound exposure. Right after the

Table 1
Relevant acoustic parameters of the four sound treatments: pulse repetition interval (PRI), pulse repetition rate (PRR), exposure duration, average zero-to-peak sound pressure level ($SPL_{z\text{-}p}$), average single-strike sound exposure level (SEL_{ss}), number of pulse and average cumulative sound exposure level (SEL_{cum}).

Treatment no	PRI (s)	PRR (s^{-1})	Duration (min)	Avg $SPL_{z\text{-}p}$	Avg SEL_{ss}	Pulse no	Avg SEL_{cum}
1	0.5	2.00	60	158	140	7200	179
2	1.0	1.00	60	158	140	3600	176
3	2.0	0.50	60	158	140	1800	173
4	4.0	0.25	60	158	140	900	170