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## SOS Bass project: acute and chronic effect of vessel noise disturbance on farmed fish

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### ABSTRACT

It is widely recognized that the marine environment is impacted by noise pollution generated by human activities. Maritime traffic is the primary source of diffuse broadband noise in the marine environment, including aquaculture systems, and can have negative effects on marine organisms. Here, we present the activities carried out and in progress within the SOS Bass project (PNRR M4C2 Investment 1.1 Research Projects of National Relevance - PRIN, funded by the European Union – NextGenerationEU), which aims to address this knowledge gap using a key species of the Mediterranean and aquaculture: the European seabass. The project began with the monitoring of the soundscape in an area of the northern Adriatic Sea and is continuing with the exposure of the animals to different recorded boat noise signatures to investigate the effects of acute and chronic exposure in terms of morphological and sensory damage, stress, and behavior. Based on the preliminary results of acute tank-based exposure experiments, the study will proceed by focusing on chronic sound exposure within offshore cages to evaluate the specimens' growth, immunity, stress response, reproduction, and behavior. Finally, potential measures to mitigate the impact of maritime traffic noise on the marine environment will be assessed, and a mitigation plan will be proposed.

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### 1. INTRODUCTION

Underwater noise pollution is an emerging environmental concern, primarily driven by human activities such as maritime traffic, industrial operations, and aquaculture. Among these, shipping traffic is the dominant source of diffuse broadband noise and is mainly due to the propeller even though other sources onboard, such as the engines add a significant contribution on it [1]. Shipping noise can interfere with marine organisms' sensory perception, communication, and overall well-being.

In general, exposure to ship noise can produce a wide range of harmful effects on invertebrates, fishes and marine mammals, going from behavioral modifications to physiological and auditory effects [2-10]. The highest intensities of noise generated by vessels usually fall within a frequency range which has been shown to be a potential threat to fish, because most audiograms of marine fish species indicate that their greatest auditory sensitivity falls within this range [11]. Exposure to ship noise pollution can elicit behavioral responses and increase physiological stress in fish [12-15] and appears to mask acoustic communication [16] with effects on foraging, navigation and reproduction [17]. Despite increasing awareness of its potential ecological consequences, the impacts of noise pollution on marine species, particularly those of commercial and ecological importance, remain poorly understood.

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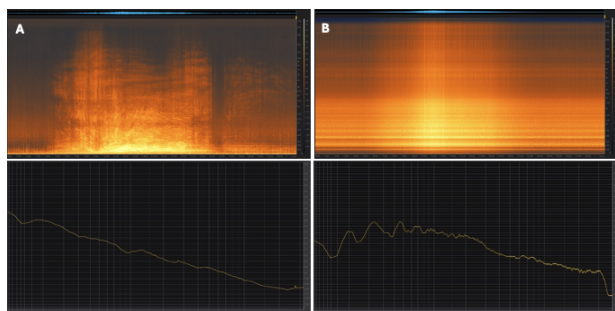
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the European Union – NextGenerationEU) aims to bridge this knowledge gap by investigating the effects of noise exposure on a key species in Mediterranean aquaculture: the European seabass (*Dicentrarchus labrax*). By combining field monitoring and controlled exposure experiments, the study assesses both acute and chronic noise effects on seabass morphology, sensory systems, stress response, behavior, growth, immunity, and reproduction. Furthermore, the project seeks to identify potential mitigation strategies to minimize the impact of maritime noise on marine ecosystems and sustainable aquaculture.

## 2. ACOUSTIC DATA RECORDING AND ANALYSIS

This study investigates the underwater acoustic environment within a seabass aquaculture farm located in Duino (Trieste, Italy). Acoustic recordings were conducted in the framework of the SOS-BASS PRIN 2022 project using an autonomous acoustic recorder model SYLENCE LP 440-DP-P-S (RTSYS - Caudan, France) deployed at the center of the fish farm, a few meters below the surface, continuously collecting data from mid-December 2023 to early July 2024 at a 64 kHz sampling rate and 16 bit of resolution.

From the acoustic recordings collected at sea, two 30-minute boat noise tracks were selected: (A) the noise from a boat considered "familiar" to the animals, as it was the same vessel that regularly visited the sea cages to deliver food; and (B) the noise from a boat that passed near the farm only once, representing an "unfamiliar" sound for the animals (see Fig. 1).



**Figure 1.** Spectrograms (FFT: 32768; Window: Hann; Frequency scale: Mel; Sampling rate: 64000 Hz) and Root Mean

Square (RMS) of shipping noise tracks from Familiar (A) and Unfamiliar (B) boats.

## 3. EXPERIMENTAL PROTOCOL

Based on these two boat noise categories and the playback modalities, four acoustic stimuli were defined for animal exposure:

1. **Familiar boat noise** played in a progressive manner (approach, proximity, and departure) with a track duration of 30 minutes;
2. **Familiar boat noise** played in an abrupt manner (maximum intensity without approach and departure phases) with a track duration of 10 minutes;
3. **Unfamiliar boat noise** played in a progressive manner (approach, proximity, and departure) with a track duration of 30 minutes;
4. **Unfamiliar boat noise** played in an abrupt manner (maximum intensity without approach and departure phases) with a track duration of 10 minutes.

A fifth condition, characterized by the background noise of fish farm's origin, will be used as Control.

### 3.1 Acute noise exposure

The acute noise exposure will be performed in 300-liter capacity tanks. For each experimental condition (experimental playback) and for the control group (background noise), 5 animals will be tested for a total of 75 fish (5 fish per tank per 3 replicates per 5 conditions). In each trial, fish will be tagged by elastomers to enable individual recognition, transferred to the exposure tank and will be exposed to one hour of experimental playback after 7 days of acclimation. A UW30 underwater speaker (Lubell Labs Inc., Columbus, OH, USA) will be placed on one side of the tank. The speaker will be connected to a Channel Low Impedance Amplifier that is in turn connected to the stereo output of a PC. Files of the vessel noise treatment will be included in a playlist and randomly projected in the "loop mode" for the entire experimental period.

Noise levels in the tank during the exposure will be monitored in terms of sound pressure by means of a calibrated hydrophone to provide the effective soundscape experienced by fish.

The effects of 1 h continuous noise exposure on fish will be evaluated in terms of morphological damage to the auditory



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system, stress response and behavior. During the observation times, the behavior of animals will be recorded by cameras located above the tank to check the initial responses to the stimulus (sudden and short-term changes in the speed and direction of swimming, also called "alarm responses"), opercular beats (ventilation rate) and swimming patterns (speed, direction, cohesion of the group, distance from the sound source). Alarm responses and ventilation data collection will be performed by trained observers on videos using the software Observer XT® (Noldus, The Netherlands), whereas the parameters of motion (distance moved, mobility state, movement and velocity) will be analyzed automatically with EthoVision 13.0 software (Noldus, The Netherlands).

## 3.2 Chronic Noise exposure

The main aim of this chronic noise exposure is to assess the mid-and long-term impact of the acoustic disturbance related to boat noise on farmed European sea bass. In particular, the results of acute noise exposure will be used to select the sounds among the four stimuli adopted. The chronic noise exposure will take place in the aquaculture farm of Valle Ca' Zuliani Società Agricola S.r.l. located in the study area. In the aquaculture site, three off-shore cages will be adopted as testing samples and the speaker projecting the sound will be set equidistant from them so that the sound propagation will be homogeneous throughout the testing area. A calibrated hydrophone will record the projected sounds in proximity of each cage to have the exact soundscape experimented by fish to be subsequently analyzed with a dedicated acoustic signal analysis software. Daily exposure duration will be planned for the selected sound and will last 4 months. Behavioral, physiological, molecular and morphological data will be collected at specific time points during the 150 days, starting 30 days before experimental noise exposure (Control phase), and continuing for the 120 days following the animal's exposure to playbacks (Testing phase) to assess medium- and long-term effects. During the Testing phase, the playbacks will be repeated discontinuously in order to simulate realistic exposure to boat and ship traffic. At predefined time points (every 30 days), a representative number of fish (N=12 per 3 replicates/cages) will be sacrificed and sampled for further analysis. Sampling time points will be: T0 (start of the Control phase), T1 (end of the Control phase - start of the Testing phase), T2 (at 30 days of exposure), T3 (at 60 days of exposure), T4 (at 90 days of exposure) and T5 (at 120 days of exposure – end of the Testing phase). The parameters of primary, secondary and tertiary stress response in blood, tissues and organism

levels will be measured at the time-points of the Control and Testing phases.

Behavioral observations will be performed on videos and scheduled in the 24 hours preceding the sampling of animals. Due to the inability to standardize the background for automatic parameter collection in off-shore cages, the collection of behavioral data in this WP will be performed by the trained observers.

## 4. MITIGATION STRATEGY FOR NOISE IMPACT

Based on the results of acute and chronic exposition of fish to shipping noises, potential mitigation measures to reduce the impact of noise on European sea bass in real environmental conditions will be investigated. These measures may target individual noise sources, particularly ship propellers, or address overall vessel traffic. The study will assess the benefits of propeller redesign and optimization in reducing noise emissions, considering the species' specific sensitivity. Additionally, broader traffic management strategies will be explored. Using Automatic Identification System (AIS) data, a noise propagation model will simulate ship-generated noise within the 100–1000 Hz range, incorporating environmental parameters such as bathymetry and sound speed profile. The resulting noise maps will be integrated with species distribution data to evaluate the impact of mitigation strategies, including traffic deviations and speed reductions, on a large scale. The outcomes will support the identification of effective noise mitigation tools and the development of a comprehensive mitigation plan.

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