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Impact of Maritime Traffic Noise on Juvenile *Dicentrarchus labrax*

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ABSTRACT

Human activities in marine environments contribute to increasing noise pollution, affecting various aquatic species at behavioral, physical and physiological levels. In aquaculture, a rapidly expanding sector, understanding the effects of noise on farmed species is essential, especially for individuals in their early growth stages, as they are more vulnerable to environmental stimuli. This study assessed the short-term effects of maritime traffic noise on the behavior, growth, and biochemical and molecular responses of juvenile *Dicentrarchus labrax*, analyzing different exposure times. The acoustic emissions consisted of recordings of noise produced by four fishing boats operating at 10-15 meters, played randomly with silent intervals ranging from 10 to 30 seconds. Preliminary results indicate significant effects on motility, group cohesion, and cellular and molecular responses. Additionally, a decrease in the growth of exposed individuals was observed. These findings provide valuable insights for managing acoustic emissions in aquaculture facilities, contributing to the improvement of animal welfare.

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1. INTRODUCTION

Chemical pollution, overfishing and maritime traffic have been identified as pressures on marine biodiversity [1]. However, in recent years, noise pollution has gained importance. The increase of the antropogenic noise, caused by human activities such as maritime traffic, infrastructure construction and offshore energy generation, negatively impacts marine ecosystems.

To mitigate the effects, new laws and European guidelines establishes a protection framework that includes underwater noise among the descriptors of the environmental status of the marine environment. Also, international agreements have driven research to classify and mitigates noise sources [2].

Ocean noise originates from natural and anthropogenic sources, classified into low frequency (10-500Hz), mid frequency (500-25 kHz) and high frequency (>25 kHz) bands [3].

Anthropogenic sources mainly affects low frequencies and propagate over long distances [4]. Natural noise includes physical and biological phenomena, such as sounds produced by fish, mammals and crustaceans [5].

Both chronic and acute anthropogenic noise can produce detectable effects on intraspecific communication, vital processes, physiology, behavioral patterns, health status and survival [6]. In species of fish, noise can impacts their hearing ability, as well as their capacity to forage for food and detect predators [7].

The European seabass (*Dicentrarchus labrax*) has been used as a species of interest due to its commercial importance in the Mediterranean. It inhabits shallow waters, tolerates wide temperature and salinity variations, and is an active predator.

2. PROCEDURES

The experiment was conducted at the facilities of the Universidad Politécnica de Valencia in the Port of Gandia, using 348 juvenile *Dicentrarchus labrax* from an aquaculture farm in Burriana. The fish were placed in tanks (147 cm x 118 cm x 89 cm) filled with water from the port, previously treated. Heaters and filters were used to maintain optimal conditions.

2.1. Acoustic set up

For the acoustic study, it was used a system of a DAQ card, an amplifier and a speaker calibrated with a hydrophone. Recordings of fishing boats at 10-15 meters were used, played randomly with silent intervals of 10 to 30 seconds.

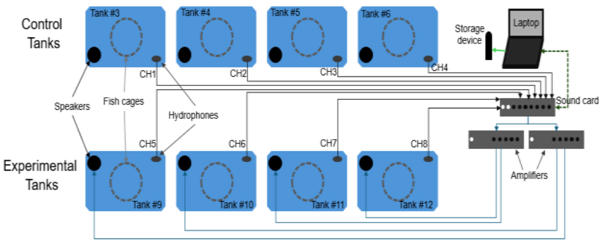

Figure 1. Acoustic set up

2.2. Experimental plan

The fish were divided into control and experimental groups, with three sampling replicates conducted after different exposure times to noise (30 minutes, 3 hours, 24 hours, 7 days). An acclimatization period was carried out between the sampling.

2.3. Biological sampling

It was measure the weight and length, with samples preserved in RNAlater or dry at -20ºC. Growth was analyzed by comparing weight and length variables in the different groups.

2.4. Behavioral analysis

To analyze behavior, video recordings were processed using Python OpenCV (Open Source Computer Vision Library), dividing the frames into 5 cm grid squares. The videos were also reviewed using Windows Media Player Classic (Microsoft Corporation, Redmond; Washington). For motility a 10 second video segment was analyzed every 6 minutes, counting the number of squares crossed by each fish. This value was converted into swimming speed (cm/s) based on the square size (25 cm^2) and analysis time (10 s). The cohesion of the group was evaluated by counting the numbers of squares occupied by the fish every 30 seconds.

2.5. Statistical Analysis

Included a normality test (Shapiro-Wilk) and group comparisons using the Mann-Whitney U test to assess significant differences in motility and group cohesion.

2.6. Cellular and molecular analysis

The levels of cortisol, heat shock protein 70, superoxide dismutase, glutathione peroxidase and adrenocorticotropic hormone were measured using the Elisa kit, while Real-Time PCR was performed on RNAlater-preserved samples.

Total RNA was extracted with the Trizol Plus Purification Kit, quantified with Nanodrop and converted to cDNA using the High-Capacity cDNA Reverse Transcription Kit. Gene expressions were analyzed by RT-PCR and using actin as the reference gene.

3. RESULTS

A total of 264 fish were analyzed, recording their weight (g) and length (cm) at different sampling times (30 minutes, 3 h, 24 h, 7 days). The data showed in Tab.1. that the weight and length were similar in the first three periods but increased significantly by the seventh day (weight: +1 g;

length: +1 cm). The standard deviation was lower at 7 days, indicating greater consistency in the measurements.

Table 1. Weight and length for each sampling time

	g	cm
30'	1,90 ± 0,18	5,51 ± 0,20
3h	1,87 ± 0,19	5,51 ± 0,24
24h	1,79 ± 0,13	5,46 ± 0,14
7d	2,60 ± 0,11	6,18 ± 0,03

In the motility analysis, preliminary results showed greater variability in the control group. Some sampling times showed significant differences in the mean, but not in the standard deviation.

The graphical comparison Fig. 2. showed that both groups increased their motility over time, although with different patterns. The control group had a more abrupt increase at the beginning and a drop at the end, while the experimental group showed a more consistent growth pattern.

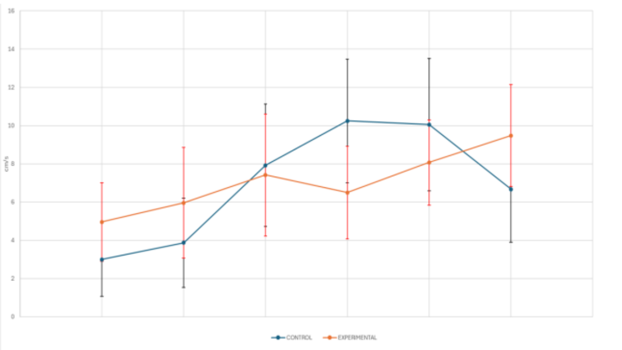

Figure 2. Line chart with error bars for the motility of the fish

Regarding biochemical analysis, preliminary results showed an increase of some parameters within 30 minutes of noise exposure then returned to normal after 7 days.

Cortisol levels increased significantly after the onset of acoustic stress, reaching their peak at 3 hours. In contrast, the activity of superoxide dismutase (SOD) and glutathione peroxidase (GPx), as well as the expression of heat shock proteins (HSPs), peaked within the first 30 minutes. However, after 7 days of exposure, all measured parameters returned to levels comparable to the control group, suggesting a physiological adaptation or recovery from the acoustic stress.

Molecular evaluations were performed on the whole fish and on each individual. Gene expression showed a modulated trend over time and was different for each gene analyzed; therefore, transcriptomic analysis will be performed.

4. DISCUSSION

The results indicate no significant differences in fish growth during the first 24 hours. However, after seven days, growth in both weight and length became significant, with an average increase of nearly 1 cm and 1 g.

Previous studies have linked acoustic stimuli to growth changes, with mixed results [8]. More pronounced effects have been observed at early growth stages, such as larval phase [9], while some research suggest that growth impairment disappears in the medium term [10]. These findings highlight the need for longer duration studies.

Motility analysis showed a general increase over time in both groups, but the control group exhibited a sharper initial rise, while the experimental group showed a more gradual increase. The variability in this parameter suggests behavioral differences, with the control group displaying greater fluctuations across the sampling periods. As for the results of group cohesion, they are still under analysis.

Overall, significant differences exist between control and experimental fish, but more data is required for definitive conclusions.

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