



In situ behavioural responses to boat noise exposure of *Gobius cruentatus* (Gmelin, 1789; fam. Gobiidae) and *Chromis chromis* (Linnaeus, 1758; fam. Pomacentridae) living in a Marine Protected Area

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ABSTRACT

The short-term behavioural effects of two types of boat noise were tested on *Gobius cruentatus* and *Chromis chromis*, i.e. one permanently and one temporarily benthic vocal fish species living inside the WWF-Natural Miramare Marine Reserve (Northern Adriatic Sea, Italy). The underwater noises produced by a 26-m tourist ferry and a 5-m fiberglass boat were recorded inside the core zone of the reserve. Each type of boat noise was subsequently played back *in situ* to 10 animals *per species* (*C. chromis* males caring their nests or *G. cruentatus* in their shelters).

The 1/3 octave spectra of recorded sound pressure levels were compared to the underwater ambient noise level and to sound pressure level measured at the hearing threshold of the two species. The boat noise levels have been calculated in terms of particle acceleration for both field measurements and *in situ* playback projections and subsequently compared to the available measured values of particle acceleration at the hearing threshold.

The animals were free to move in all directions during the whole experimental session. The behaviour of each fish was videotaped by an underwater camera for a total of 10 min (5 min before and 5 min during the noise playback). No short-term behavioural reaction (aversion) was observed in any of the specimen of the two species during the playback of the recorded noises, therefore suggesting no impact. However a time-budget analysis revealed a significant change in the total time spent in caring their nests (*C. chromis*) or inside their shelters (*G. cruentatus*). This result highlighted how analyzing fish reaction on a short-term might underestimate the effects of noise disturbance and indicated that the overall fish behaviour should be considered to assess noise impact.

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1. Introduction

Worldwide concern about the impact of noise pollution on aquatic fauna is growing in these years. There is an increasing amount of scientific evidence that anthropogenic noise can harm marine species (Tyack, 2008). While noise pollution has been recognised to be steadily growing in the world's oceans (Andrew et al., 2002), this phenomenon is still largely unmonitored in coastal areas. A major source of low-frequency noise (under 1000 Hz) in marine species living in highly anthropized coastal areas comes from boats and vessels, since their number, distribution and mobility are very high (Greene and Moore, 1995; Richardson and Würsig, 1997).

Boat noise represents a chronic source of harassment (Haviland-Howell et al., 2007) for fish species (Popper, 2003), whose communication for inter- and intra-sexual selection is mainly based on low-frequency sound signals (Ladich and Myrberg, 2006; Myrberg and Lugli, 2006). It has been recently shown that boat noise may induce endocrine stress response (Wysocki et al., 2006), as well as diminish hearing ability and mask intra-specific relevant signals in exposed fish species (Scholik and Yan, 2002; Amoser et al., 2004; Vasconcelos et al., 2007; Codarin et al., 2009). In addition, boat and vessel noises have the capacity to provoke short-term changes in the spatial position and group structure of pelagic fish in the water column, as shown by many studies carried out since the 1960s (for example, Buerkle, 1974; Olsen et al., 1983; Schwarz and Greer, 1984; Engås et al., 1995; Soria et al., 1996; Vabø et al., 2002; Mitson and Knudsen, 2003; Ona et al., 2007; Sarà et al., 2007). The most common boat-induced behavioural changes in fish include the temporary cessation of activities, alarm response, flight reaction or the so-called 'startle' response, i.e. a powerful flexion of the body followed by a few seconds of faster

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swimming (Boussard, 1981). In many species, fish behaviour is affected by noise only when a certain threshold in pressure level is reached. Very often, the previously mentioned short-term changes in swimming speed have been used to fix the threshold of fish behavioural reaction to human noise (Kastelein et al., 2008a), but such noise-response studies on marine fish are rare (Akamatsu et al., 1996) and they show marked differences in the reaction of various species, depending on the threshold levels of the noise frequencies, the threshold levels at which a reaction occurs varying *per* frequency for each species (Kastelein et al., 2008b). The relationship between the strength of short-term responses and the underlying sensitivity of wildlife is unlikely to be straightforward (Gill et al., 2001) and fish reactions depend not only on the properties of noise but also on the individual context (e.g. location, temperature, physiological state, age, body size, etc.). As result, much more information is still needed to understand the behavioural consequences of anthropogenic noise exposure (Popper et al., 2004).

Interestingly, the application of a time-budget analysis has recently proved to be a useful tool for assessing human disturbance in several cetacean species (e.g., Williams et al., 2006; Hodgson and Marsh, 2007; Dans et al., 2008; Stockin et al., 2008). This technique has never been applied to fish species so far, therefore, the aim of this study was (1) to record, inside a core zone of a coastal reserve, the noise produced by a tourist ferry and a fiberglass boat moving along and inside the Marine Protected Area (MPA); (2) to field-test, through the time-budget method, short-term effects of both boat noise types on a permanently and a temporarily benthic soniferous fish species (*Gobius cruentatus* and *Chromis chromis*) living inside the MPA.

Investigating the impact of boat noise on target fish species is particularly relevant for coastal MPAs, which are biologically rich locations in highly populated regions and deserve protection from anthropogenic pollutants. Managers of MPAs have recently begun to study noise (Agardy et al., 2007; Haren, 2007) but far too little is known about animals hearing capacity, behaviour and ecology to set a standard or apply an exposure limit with confidence (Popper and Løkkeborg, 2008). This high level of uncertainty underlines the need for local assessment in noise pollution as well as a precautionary principle as management rule for sensitive areas (Horowitz and Jasny, 2007).

2. Methods

2.1. Study area

The field-work has been run at the WWF-Miramare Natural Marine Reserve, an UNESCO-MAB Biosphere Reserve located in the Gulf of Trieste (Northern Adriatic Sea, Italy) at 45°42'08" N and 13°42'42" E. The area is divided in a core (30 ha) and in a buffer zone where the maximum depth reaches 18 m. The level of human presence around Miramare MPA is extremely high compared to more remote Mediterranean MPAs. The site is less than 8 km away from the city of Trieste, an important seaport with more than 48 million tons of ship traffic per year. The site is also very close to a tourist port characterized by high recreational boat traffic. However, the coastline of the reserve (1700 m) and its offshore area (120 ha) are densely populated by several fish species (Guidetti et al., 2005), most of which spawn during summer. This makes the Miramare Reserve an important seasonal nursery area in the North Adriatic Sea. A recent study, which assessed the number of vessels moving in the Gulf of Trieste and their distance from the reserve (Codarin et al., 2008), showed that tourist ferry boats that connect different ports along the local coastline during the summer are a possible source of noise disturbance for local fish species, due to their frequency (4 times a day) and their route, which is very close to the limit of the core zone of the MPA. In addition, a fiberglass boat used by the MPA staff for moving inside the area during research and educational activities has been labelled as another source of annoyance.

2.2. Target species

The red mouthed goby, *G. cruentatus*, is a small benthic member of the Family Gobiidae and is common in Mediterranean Sea and in Western Atlantic Ocean. During the year, it lives in rock crevices (Wilkins and Myers, 1993), defending itself from intruders using visual and acoustic displays (Picciulin et al., 2006). Acoustic displays consist in four different types of sounds, ranging in peak frequency from 82 Hz to 185 Hz (Sebastianutto et al., 2008). Recently the species' audiogram, as well as the audiogram of *C. chromis*, has been described in terms of both sound pressure and particle acceleration (Wysocki et al., 2009). The hearing range of *G. cruentatus* reaches 700 Hz, with highest sensitivity to sound pressure at 300 Hz and highest sensitivity to sound particle acceleration at 200 Hz (Wysocki et al., 2009). Unlike other goby species, *G. cruentatus* has a swim bladder (Gil et al., 2002).

The Mediterranean damselfish, *C. chromis*, is a common small fish that lives in shoals in the Mediterranean Sea, between 3 and 30 m deep. From June to September males synchronously establish territories, prepare nests and court females through visual displays (Abel, 1961) and acoustic signals, i.e. broadband pulses, called "pops," peaking at about 400 Hz (Picciulin et al., 2002). Females lay demersal eggs that are guarded and fanned by males until hatching. When hatching of the eggs is concluded, males leave the nests and rejoin the feeding school; males remain on their territory for about 10 days. *C. chromis*' audiogram reaches an upper frequency limit of 600 Hz and shows highest sensitivity to both particle acceleration and sound pressure at 200 Hz (Wysocki et al., 2009).

In the study area, both species live and reproduce in waters between 3 and 7 m deep on a 150-m coastal rocky reef located inside the core zone of the Natural Marine Reserve of Miramare. Being as far as possible from the limits of the core area, this reef can be considered the most protected area. It can be used as a reference point for boat noise recordings, assuming that if a disturbance from noise recorded at this point is noticeable, a much larger impact of the stimulus can be expected along the whole MPA.

2.3. Boat noise recordings

The noise emissions of a 26-m tourist ferry (TF) with inboard diesel engine moving at 6 kn along the perimeter of the core zone of the Miramare MPA and a 5-m fiberglass boat (FB) with 40 HP outboard engine moving at 15 kn were recorded on the 6th June 2004 during daytime in the coastal rocky reef where fish density of target species was high (Fig. 1). A calibrated Reson TC4032 hydrophone (sensitivity – 170 dB re 1 V/ μ Pa; frequency range: 5 Hz–120 kHz) was placed underwater, 4 m deep from a boat (bottom depth: 8 m) and connected to a Pioneer DC-88 DAT recorder (sample rate 44.1 kHz, 16-bit) operating on batteries. During the recordings, water temperature – equal to 18 °C – was measured at the same depth using a multiparametric 316 CTD-Idronaut probe. The distances of the hydrophone between TF and FB were 82 m and 1 m, respectively. Distances were calculated by hand-held GPS. The recording conditions were: sea state 0–1 (Douglas scale), wind speed 7–15 km/h, and few clouds (5% clouds at maximum). Each recording lasted 60 s. As a comparison, 60-second-long samples of the sea ambient noise (SAN) were collected at the same site and depth when no boats were moving in a range of 10 nautical miles from the recording point.

Samples of 25 s from SAN, FB and TB were considered for the analysis, the boat samples included the highest amplitude value of the noise. The noises were analysed looking at instantaneous sound pressure levels (SPL, L-weighted, 20 Hz to 20 kHz, RMS fast) with Spectra RTA (Sound Technology) spectral analyser, operating in 1/3 octave bands. Each recording was previously calibrated with a signal of 100 mV RMS at 1 kHz recorded at the start of each tape. The equivalent continuous SPL (L_{Leq}) was calculated averaging the instantaneous SPLs values over 25 s.

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