Bioacoustics
The International Journal of Animal Sound and its Recording, 2011, Vol. 20, pp. 117–136

EFFECTS OF AQUARIUM AND POND NOISE ON HEARING SENSITIVITY IN AN OTOPHYSINE FISH

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ABSTRACT

Several studies on fishes have shown that behaviour and auditory sensitivity are often affected by underwater noise. The current investigation concentrates on noise encountered by fish kept for leisure in aquaria and ponds. Noise spectra showed that all aquarium filters measured created a high amount of low-frequency noise, while the water outflow above the surface created additional high-frequency noise components. Audiograms of the Goldfish *Carassius auratus*, a species possessing hearing specializations, were determined between 0.1 and 4kHz using the non-invasive auditory evoked potential (AEP) recording technique. The amount of masking was determined in the presence of four different noise-types (broadband $L_{Leq,\ 1min}$): aquaria with external filter with outflow above the water surface (119dB re 1 µPa), external filter with outflow below the water surface (115dB), internal filter with outflow below the water surface (114dB), and an unfiltered pond (95dB). The goldfish's hearing was masked by all filter noise types and most affected at 0.1 and 0.3kHz by the external filter noise (threshold shifts of 15-19dB). Pond noise had no effect on the hearing threshold. The results indicate that fish with hearing specializations are considerably masked under common holding conditions found in aquaria but probably not in ponds. Thus, using a quieter filter setup with a quiet outflow might help to improve holding conditions in aquaria without compromising aeration of the water.

Keywords: Sound pressure level; aquarium noise; hearing; fish; auditory evoked potentials, masking

INTRODUCTION

Sound is an important carrier of information for communication and orientation in aquatic environments because it propagates five times faster than in air and is not attenuated as quickly as other signals. There are numerous noise sources in the underwater environment, and some information is available about the effects of noise on hearing thresholds of fish species having different auditory capacities (Amoser & Ladich 2005; Ladich 2008; Codarin *et al.* 2009; Popper & Hastings 2009; Slabbekoorn *et al.* 2010).

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Fish live in an environment where the acoustic background varies greatly due to the influence of currents, waves, the prevailing weather conditions and other factors. This ambient noise is the ubiquitous acoustic background consisting of abiotic (wind, waves, rain, surf) and biotic (animal vocalizations, feeding sounds) sources (Hawkins & Chapman 1975; Hawkins & Myrberg 1983; Wysocki *et al.* 2007a; Amoser & Ladich, 2010). Many fish species are able to produce sounds via numerous sound generating mechanisms and communicate acoustically (Ladich & Fine 2006). Their acoustic signals have a certain stability in their amplitude, temporal and frequency characteristics. In order to extract relevant information from the auditory scene and to facilitate orientation and intraspecific acoustic communication, the fish ear should be specialized for hearing signals in different background noises (Ladich 2008; Fay 2009). Knowledge about the ability of fish to discriminate signals from noise improves our understanding of the adaptive mechanisms of this sensory organ. It also provides crucial insights into the mechanisms that process acoustic information (Sorokin 1989; Popper & Fay 1993).

Several studies on fishes showed that behaviour and auditory sensitivity can be affected by underwater noise (Myrberg 1990). Fish are exposed to a wide range of waterborne, anthropogenic noise under both natural and cultured conditions. In natural aquatic environments, such noise is generated by machinery, propulsion systems of ships and by-flow (Vasconcelos *et al.* 2007, Codarin *et al.* 2009). Other sources of sounds include air guns, aircraft, sonic booms, sonar systems, shock tests, boat repairs, underwater explosions and car traffic (Popper 2003, Popper & Hasting 2009).

An even greater amount of noise is generated in an aquaculture environment because aquaculture systems continue to intensify. Intensification requires the use of aerators, air and water pumps, tractors, harvesters, water circulation, as well as feeding and maintenance machinery. Consequently, fish in aquaculture facilities are chronically exposed to noise levels that are well within their hearing ranges (Wysocki *et al.* 2007b).

Noise exposure can variously affect fishes, including temporary hearing loss (Scholik & Yan 2001, 2002; Amoser & Ladich 2003; Smith *et al.* 2003; Popper *et al.* 2005), impaired sound detection and temporal resolution ability (Wysocki & Ladich 2005a, b), damage to the sensory epithelia of the inner ear (Hastings *et al.* 1996; McCauley *et al.* 2003), and endocrinological and physiological stress responses (Smith *et al.* 2003; Wysocki *et al.* 2006; Anderson 2009). Banner and Hyatt (1973) analyzed the effects of such noise on the eggs and larvae of two estuarine species, *Cyprinodon variegatus* and *Fundulus similis*. These authors showed that a 20 dB increase of sound level in the 40 to 1000 Hz frequency range reduced the viability of eggs and larvae in *C. variegatus*. Terhune *et al.* (1990) showed that noise levels may influence Atlantic Salmon smolting rates in tanks. There was a general tendency for smolting rates to be higher in fibreglass than in the noisier concrete tanks.

Artificial holding conditions are often noisier than natural habitats. In holding tanks, high-frequency underwater noise is produced mainly by oscillating and collapsing air bubbles, electric generators, as well as electric air and filter pumps, whereas low-frequency noise is mainly generated by water flows, ground vibrations, aquarium wall vibrations and electrical pumps (Bart *et al.* 2001; Davidson *et al.* 2007; Anderson 2009). Kratochvil & Schwammer (1997) showed that the well-being of fish in public aquaria requires minimizing noise levels.

The major goals of the present study were (1) to characterize various aquarium noise types resulting from different filtering techniques typically used in private ornamental fish keeping as compared to the ambient noise in a pond with no water-maintaining device and (2) to investigate the effects of these various noise conditions on hearing sensitivity in a species possessing well-developed hearing abilities due to accessory hearing structures (hearing specialist). An otophysine species having Weberian ossicles which connect the swimbladder to the inner ear was chosen because prior studies showed that noise had major effects on hearing in otophysines and none or only minor ones in species having no known hearing specializations such as perciforms and salmoniforms (Amoser & Ladich 2005; Wysocki & Ladich 2005a; Wysocki *et al.* 2007b). (3) A final goal of the study was to determine better acoustical holding conditions for fishes kept for leisure. The Goldfish *Carassius auratus* (Cyprinidae) was chosen because its hearing abilities have been well characterized in numerous studies.

MATERIALS AND METHODS

Animals

The test subjects were six Goldfish *Carassius auratus* (92-128 mm standard length (SL), 20-60 g body weight (BW) from a pond near Vienna. All animals were kept at 23 ± 1°C in planted aquaria whose bottoms were covered with sand, equipped with half flower pots as hiding places, filtered by external filters, and maintained at a 12L:12D cycle. The fish were fed commercially prepared pond or flake food (Tetrapond® or Tetramin®).

All experiments were performed with the permission of the Austrian Federal Ministry for Education, Science and Culture (GZ 66.006/0002-BrGT/2006).

Noise recording and sound pressure level measurements

The different noise types were recorded using a DAT recorder (Sony TCD-D100, Sony Corporation, Tokyo, Japan). Representative sound pressure level (SPL) values of lab, pond, and aquarium noise types were measured using a sound level meter (Brüel and Kjær 2238 Mediator) and a hydrophone (Brüel and Kjær 8101, Nærum, Denmark; frequency range: 1 Hz-80 kHz ±2 dB; voltage sensitivity: -184 dB re 1 V/μPa), both connected to a power supply (Brüel and Kjær 2804). For that purpose the *L*-weighted (5 Hz-20 kHz) equivalent continuous SPL (L_{Leq}) averaged over 1 min of measuring time was determined. The L_{Leq} is a measure of the averaged energy in a varying sound field and is commonly used to assess environmental noise (ISO 1996, 2003). The whole system was calibrated using a Brüel and Kjær 4229 calibrator.

Aquarium noise was recorded in an aquarium (water temperature: 23 ± 1°C) at the animal keeping facilities in the Department of Behavioural Biology at the Biocenter in Vienna. The aquarium was 1 × 0.5 × 0.5 m in size with approximate 200 l of freshwater and sand on the bottom (Fig. 1). It was placed on 2 cm of styrofoam, a 3 cm wooden board and a metal frame. The hydrophone was placed in the middle of the aquaria. An external (Eheim Ecco 2232) and an internal filter (Eheim Aquaball 2212) with variable water outflow were tested. The end of the outflow pipe of the external filter was either plugged or unplugged. The internal filter possessed an adjustable water outflow rate and a diffuser which made it possible to regulate the amount of air added to the water (aeration).

The pond is located in Prellenkirchen, southeast of Vienna (geographical position: 48.1°N, 17.0°E; altitude: 163 m above sea level), measures 32 × 22 m with an approximate depth of 1.8 m, and is populated by cyprinids. The ambient noise was a mixture of biological activity, natural water movement due to wind, and small surface waves characteristic for the summer season. The underwater noise was recorded on 29 July 2005 at two different places. It was a warm (water-temperature: 26.2°C) and slightly windy day. The hydrophone was positioned approximately 0.5 m below the surface. Before and after each noise recording, the SPL (L_{Leq}) of the ambient noise was measured and then averaged.

Lab noise was recorded in the water tub where the auditory evoked potential (AEP) recordings took place.

Noise spectra calculations

All noise recordings (sampling frequency of 44.1 kHz) were analysed using the acoustic analysis software S_TOOLS-STx 3.7 (Acoustics

Research Institute, Austrian Academy of Sciences, Vienna, Austria). Averaged sound spectra of pond, aquarium and lab noise were calculated according to Amoser *et al.* (2004) and Wysocki & Ladich (2005a).

Auditory evoked potential recordings

The AEP recording protocol followed was developed by Kenyon *et al.* (1998) and modified by Wysocki & Ladich (2005a, b). During the experiments, the fish were mildly immobilized with Flaxedil (gallamine triethiodide; Sigma-Aldrich, Vienna, Austria). The dosage used was 0.88 ± 0.25 $\mu g\ g^{-1}$. This dosage allowed the fishes to retain slight opercular movements during the experiments but without significant interference of myogenic noise. Test subjects were secured in a bowl-shaped plastic tub (diameter: 33 cm, water depth: 13 cm, 1.5 cm layer of sand) lined on the inside with acoustically absorbent material (air-filled packing wrap) in order to reduce resonances and reverberations (for the illustration of the effect, see Fig. 1 in Wysocki & Ladich 2002). Fishes were positioned below the water surface (except for the contacting points of the electrodes, which were up to 1 mm above the surface) in the centre of the plastic tub (Ladich & Wysocki 2009). A respiration pipette was inserted into the fish's mouth and respiration was achieved through a simple temperature-controlled (23 ± 1°C), gravity-fed water system. The AEPs were recorded by using silver wire electrodes (0.38 mm diameter) pressed firmly against the skin. The portion of the head above the water surface was covered by a small piece of Kimwipes® tissue paper to keep it moist and to ensure proper contact during experiments. The recording electrode was placed in the midline of the skull over the region of the medulla and the reference electrode cranially between the nares. Shielded electrode leads were attached to the differential input of an a.c. pre-amplifier (Grass P55C, Grass Instruments, West Warwick, RI, USA). A ground electrode was placed in the water. The plastic tub was positioned on an air table (TMC Micro-g® 63-540, Technical Manufacturing Corporation, Peabody, MA, USA), which rested on a vibration-isolated concrete plate. The entire setup was enclosed in a walk-in soundproof room, which was constructed as a Faraday cage (interior dimensions: 3.2 m × 3.2 m × 2.4 m) (Figure 1).

Both, sound stimulus presentation and AEP waveform recordings were accomplished using a Tucker-Davis Technologies (TDT, Gainesville, FL, USA) modular rack-mount system (TDT System 3) controlled by a Pentium PC containing a TDT digital processing board and running TDT BioSig RP Software.

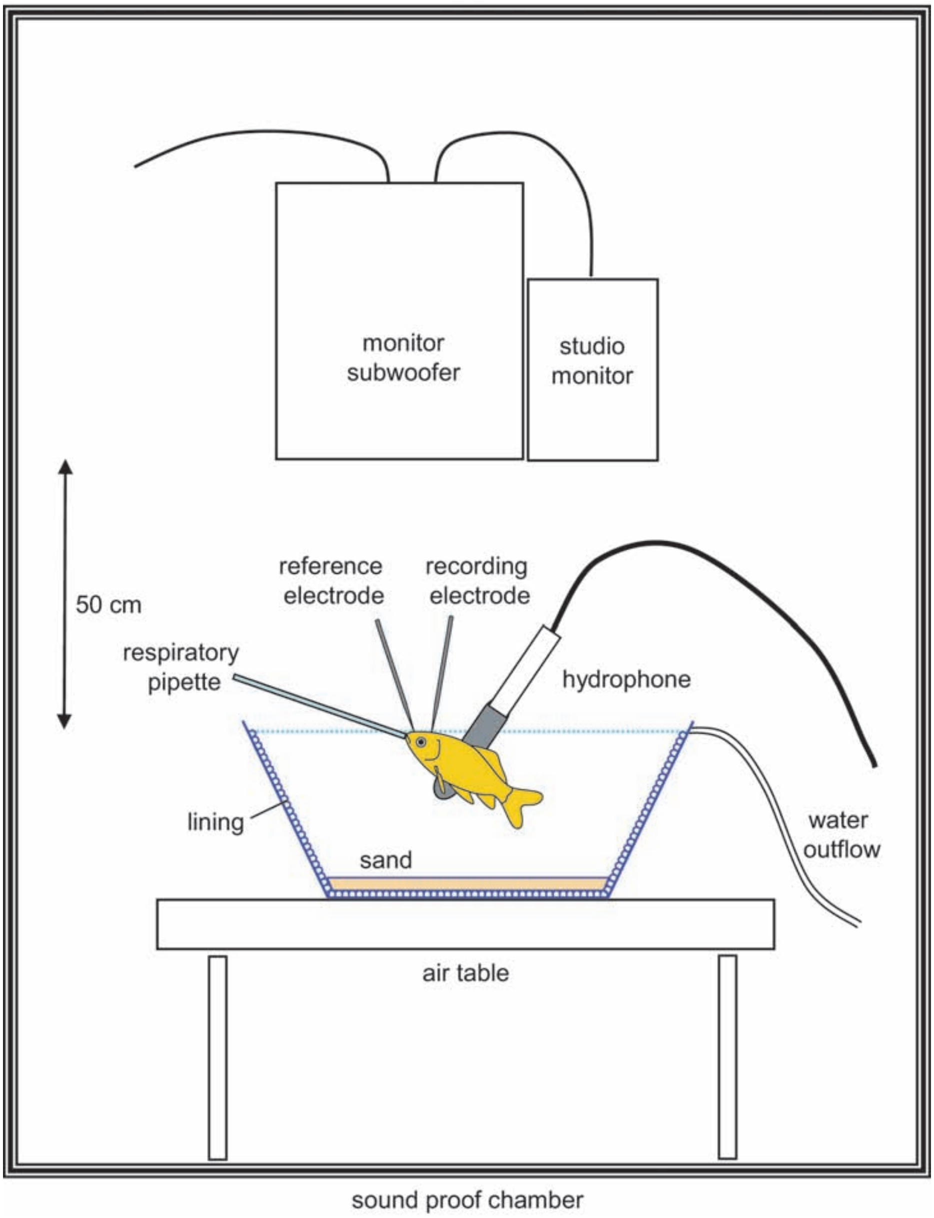

Figure 1. AEP recording and sound (noise) presentation setup showing both speakers (monitor subwoofer and studio monitor), the plastic tub lined on the inside with acoustically absorbent material, and the fish position. The arrow indicates the distance between the water surface (stippled line) and the speakers (modified from Ladich and Wysocki 2009).

Sound stimuli

Sound stimuli waveforms were created using TDT SigGen RP software and fed to two speakers (Fostex Professonial Monitor Subwoofer PM-0.5 Sub and Professional Studio Monitor PM-0.5 MKII, Fostex Corporation, Tokyo, Japan) mounted 0.5 m above test subjects in the air (Figure 1). Sound stimuli consisted of tone bursts presented at a repetition rate of 21 per second. Hearing thresholds were determined at frequencies of 0.1, 0.3, 0.5, 0.8, 1, 2, 3 and 4 kHz. Frequencies were presented in a random order under normal laboratory conditions, as well as in the presence of continuous masking noise. The duration of sound stimuli increased from two cycles at 0.1 and 0.2 kHz, up to eight cycles at 4 kHz. Rise and fall times were one cycle at 0.1 and 0.2 kHz, and two cycles at all other frequencies. All bursts were gated using a Blackman window.

For each test condition, stimuli were presented at opposite polarities (180° phase shifted), and the corresponding AEPs averaged by the BioSig RP software in order to eliminate stimulus artefacts. The sound pressure level (SPL) of the tone bursts was reduced in 4 dB steps until the AEP waveform was no longer apparent. The lowest SPL for which a repeatable AEP trace could be obtained, as determined by overlaying replicate traces, was considered the threshold (Kenyon *et al.* 1998). All hearing thresholds are given in dB re 1µPa because otophysine fish are pressure sensitive due the connection of the gas-filled swimbladder to the inner ear (Ladich & Popper 2004; Popper & Fay in press).

A hydrophone (Brüel and Kjær 8101, Nærum, Denmark; frequency range: 1 Hz - 80 kHz ± 2dB; voltage sensitivity: -184 dB re 1 V μPa^{-1}) was placed close to the right side of the animals (2 cm apart) in order to determine absolute SPL values underwater in the immediate vicinity to the subjects.

Masking noise

For playback of aquarium and pond noise during AEP recordings, 30 s of three aquarium noise recordings with the hydrophone in the middle of the aquarium were chosen: a) an external filter with vertical outflow (end of pipe plugged) 3 cm above the water surface (EFa), b) external filter with vertical outflow (end of pipe plugged) below the water surface (EFb) and c) with an internal filter (IF) with horizontal outflow below the water surface representing aquarium conditions (Figure 2), and one recording of the pond noise (P) representing a typical seminatural habitat of fish kept for leisure.

These recordings were played back in a continuous loop during the AEP-recordings. Noise, as well as tone bursts, were presented

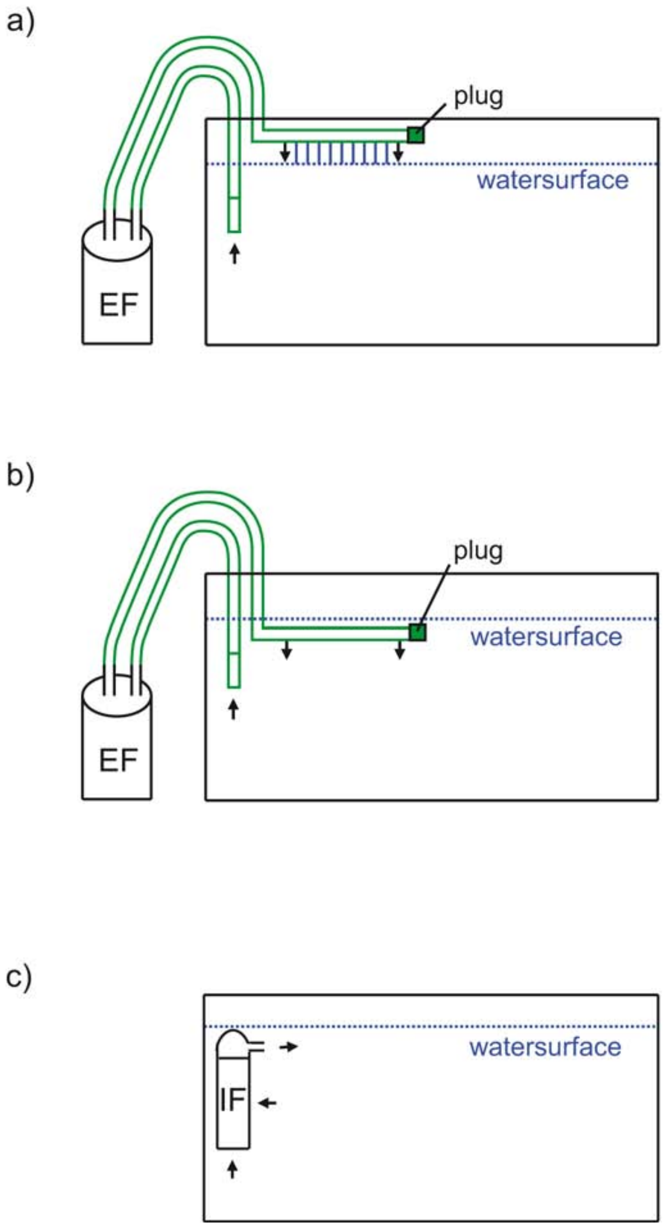

Figure 2. Filter and water flow setups: a) External filter (EFa) with outflow above water surface, b) External filter (EFb) with outflow below surface, c) Internal filter (IF) with outflow below surface. Arrows indicate water flow in different filter setups.

through two speakers (Fostex PM-0.5 Sub and PM-0.5 MKII, Fostex Corporation, Tokyo, Japan). These were positioned 0.5 m above the water surface to achieve low-frequency noise spectral amplitudes similar to those of the aquaria and pond recordings (Figure 1). Noise stimuli waveforms were generated using TDT Sig-Gen RP software sent to a 30-band equalizer (Alesis MEQ 230, Alesis Corp., Los Angeles, CA, USA) and adjusted to ensure that the spectra in the experimental tub were similar in frequency content and level to the original noise recorded in the pond and aquaria. For this purpose, the playback noises were recorded with a hydrophone (Brüel and Kjær 8101) suspended in the AEP recording tub at the position of the fish. The average equivalent continuous SPLs ($L_{Leq,\ 1min}$) measured using the same equipment as for the noise recordings in the aquaria and the pond. Absolute sound spectra of the playback noises were then calculated as described in Wysocki and Ladich (2005a) and compared to the original recordings.

Using external or internal filters with different outflow positions in relation to the water surface represents snapshots of the noise situation in aquaria, since the acoustic characteristics of filter systems tend to vary depending on the filter type used, its location relative to the aquarium and the position of the water outflow relative to the water surface. Nevertheless, the broad range of both, the level and spectral composition of the selected noise types fits our purpose to test the hearing abilities and the degree of masking in fishes kept for leisure in aquaria and ponds based on representative examples.

Statistical analysis

All audiograms obtained in the presence of the different noise types (lab-, pond- and three aquarium-noises) were compared by a two-factor analysis of variance (ANOVA) using a general linear model where one factor was masking noise and the other was frequency. The noise factor alone should indicate overall differences between masking conditions and, in combination with the factor frequency, reveal whether different tendencies exist at different frequencies of the audiograms. This was followed by Bonferroni's multiple comparison procedures to test under which noise conditions the audiograms differed from each other.

Parametric statistical tests were applied because the data were normally distributed and showed homogeneity of variances. All statistical tests were run using SPSS 15.0.

RESULTS

Diversity in noise levels and spectra

The continuous equivalent sound pressure levels ($L_{Leq,\ 1\ min}$) of aquaria and the ponds, as well as the noise spectra, differed considerably (Table 1, Figure 3).

The investigations of diverse filter types and outflow setups in aquaria revealed different noise levels. The external filter with the outflow of the plugged outlet pipe above the water surface was approximately 3 dB louder than with the underwater outflow. The SPL of the outflow of the outlet pipe with a plug at the end was up to 5.5 dB higher than without such a plug (Table 1). The SPL (L_{Leq}) of the internal filter with the outflow below the water surface increased when maximizing the aeration rate (119 dB). In the pond outside Vienna, the SPL of the two different testing sites was almost identical (95.4 dB and 94.9 dB) and about 20 dB lower than in the aquarium.

Different locations of the external filter relative to the aquarium were also investigated (Table 2). The SPL was lowest when the filter had no contact to the frame of the aquarium (111-113 dB). There was nearly no difference when the filter was located below the aquarium

TABLE 1

Mean continuous equivalent sound pressure levels ($L_{Leq,\ 1\ min}$) of different aquarium noise types and the pond at different places. The water flow and the amount of aeration can be regulated in the internal filter. EFa, external filter with outflow above surface; EFb, external filter with outflow below water surface; IF, internal filter.

Noise types	SPL (L_{Leq}) dB re 1 μPa
EFa (outflow vertical to surface; with plug)	118.9
EFa (outflow vertical to surface; without plug)	113.4
EFa (outflow 45° to surface; with plug)	116
EFa (outflow 45° to surface; without plug)	113.9
EFb (outflow below surface; with plug)	114.2
EFb (outflow below surface; without plug)	113.9
IF (max. water flow, max. aeration)	117.2
IF (max. water flow, min. aeration)	117.2
IF (min. water flow, max. aeration)	119
IF (min. water flow, min. aeration)	113.5
pond (first site)	95.4
pond (second site)	94.9

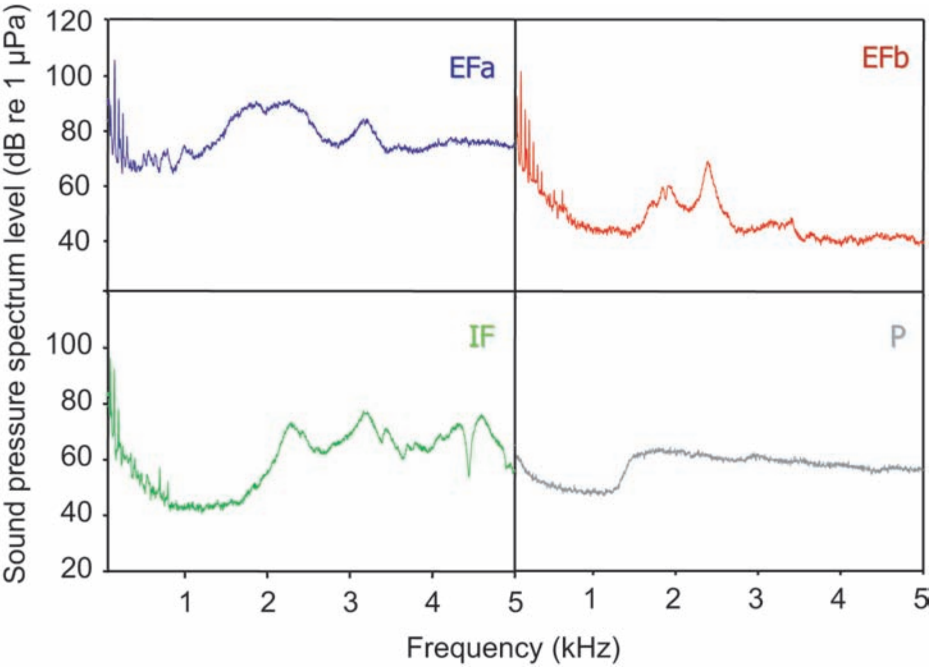

Figure 3. Sound power spectra and cepstra of the different aquarium noise types and of the pond. EFa, external filter with outflow above water surface; EFb, external filter with outflow below surface; IF, internal filter; P, pond. Note the linear frequency axis scaling in this figure and the logarithmic scaling in figure 4.

TABLE 2

Mean continuous equivalent sound pressure levels ($L_{Leq,\ 1\ min}$) of different positions of the external filter relatively to the aquarium. EFa, external filter with outflow above surface; EFb, external filter with outflow below water surface

Filter position	EFa dB re 1 μPa	EFb dB re 1 μPa
below aquarium on 2 or 5 cm of styrofoam (same frame as aquaria)	113.3	112
on a chair (no contact with aquarium)	113.1	113.1
on the floor (no contact with aquarium)	111	110.7
on a wooden board (left of aquarium)	114	112
on soft tissue (left of aquarium)	113.5	112.4

on 2 cm or 5 cm of styrofoam (113 dB) versus standing directly next to the aquarium on a wooden board or on a soft tissue (114 dB).

Four noise types were chosen to investigate the masking effect on fishes. These types are frequently encountered by fish kept for leisure: noise types of aquaria with either an external filter with a vertical outflow above the water surface (EFa) and an external filter with outflow below the water surface (EFb), an internal filter (IF) with the outflow below the water surface with minimized outflow and aeration, and the noise of the pond (P) outside Vienna representing a semi-natural habitat.

Among the four filter noise types chosen, EFa was the noisiest (119 dB), whereas the filters with the underwater outflows (EFb, IF) were quieter. Noise spectra showed that all filters created abundant low-frequency noise, but the spectral levels differed considerably at frequencies above 1.5 kHz (Figure 3). EFa showed the highest spectral levels among all aquarium noise types (> 60 dB re 1 μPa) and a major noise boost from 1 to 2.5 kHz. EFb revealed a moderate decline towards higher frequencies, but also featured an energy rise like EFa from 1.5 to 3 kHz with a peak about 2.4 kHz (68 dB re 1 μPa). IF showed a similar decline as EFb until 1.5 kHz, but then sound energy increased to 69 dB re 1 μPa. IF had, at higher frequencies, unsteady sound energy (54-70 dB re 1 μPa). The pond (P), representing a semi-natural habitat, showed lower spectral levels than the aquarium noise types in the low frequency range (<0.4 kHz), a broad noise window (about 50 dB re 1 μPa) and a quick increase in spectral level at 1.4 kHz. The pond noise had a flat, moderate decline towards higher frequencies.

Hearing under aquarium and pond noise conditions

The baseline audiogram (measured under lab noise conditions) for the goldfish showed greatest hearing sensitivity between 0.5 to 1 kHz, with hearing thresholds lower than 75 dB and a quick decline in sensitivity above 1 kHz. Comparing the baseline audiogram with the different masked audiograms by a two-factor ANOVA revealed overall significant differences between audiograms ($F_{4,200} = 2856$, $P < 0.001$) and a significant interaction between noise and frequency ($F_{28,200} = 69.4$, $P < 0.001$). This indicates different effects of noise at different frequencies of the audiogram. The Bonferroni-adjusted *post-hoc* test showed that baseline audiograms were significantly different from each other filter noise type (EFa, EFb and IF) but not from the pond noise audiogram.

Playing back pond noise had no effect on the hearing thresholds at any of the frequencies tested, but noise from aquaria had pronounced effects on auditory sensitivity as compared to measurement under lab noise (Table 3, Figure 4). In the low frequency range (0.1 and 0.3 kHz), thresholds did not differ in the presence of both EF noise types. At higher frequencies (0.8 and 1 kHz), thresholds with EFa were higher than with EFb noise. Hearing thresholds were masked by up to 20 dB (EFb at 0.1 kHz) and up to 24 dB (EFa at 0.5 kHz). In the presence of IF, the mean sensitivity at 0.1 and 0.3 kHz declined by maximally 10 dB, whereas it decreased above 1 kHz up to 13 dB. In the best hearing range of goldfish (0.5, 0.8 and 1 kHz) the amount of threshold shift compared to the worse hearing range decreased for EFb and increased for EFa and IF (Figure 5).

DISCUSSION

Diversity in noise conditions

Ambient noise in different habitats is highly diverse in terms of both noise levels and energy distribution (Wysocki *et al.* 2007a; Amoser & Ladich 2010). The natural habitats of species possessing no specializations often have relatively high ambient noise levels (Lugli & Fine 2003). SPLs in creeks and streams are usually above 110 dB re 1 µPa (L_{Leq}), whereas ambient noise levels in stagnant habitats with high percentages of species possessing hearing specializations (such as backwaters and lakes) are typically below 100 dB re 1 µPa (Wysocki *et al.* 2007a; Amoser & Ladich 2010). This is consistent with current results, where the noise level in the pond outside Vienna was 95 dB re 1 µPa $L_{Leq,\ 1\ min}$, and with data of Anderson (2009).

Human-made holding conditions are often noisier than natural habitats. A wide range of waterborne noise has been observed during

TABLE 3

Mean (± S.E.M.) hearing threshold values of *Carassius auratus* measured under the different background noise conditions. S.E.M., standard error of means; EFa, external filter with outflow above surface; EFb, external filter with outflow below water surface; Freq, frequency; IF, internal filter; P, pond.

Freq kHz	Baseline dB re 1 µPa	EFa dB re 1 µPa	EFb dB re 1 µPa	IF dB re 1 µPa	P dB re 1 µPa
0.1	87.33 ± 1.43	102.50 ± 1.86	106.67 ± 2.08	97.33 ± 1.48	87.67 ± 1.59
0.3	75.17 ± 2.30	90.67 ± 2.23	93.50 ± 2.23	82.67 ± 1.17	76.33 ± 2.33
0.5	68.00 ± 1.98	85.50 ± 1.03	77.83 ± 2.47	80.33 ± 1.50	68.33 ± 2.38
0.8	63.00 ± 2.25	86.50 ± 2.08	71.50 ± 1.91	81.50 ± 1.54	63.33 ± 2.47
1	69.17 ± 2.06	87.83 ± 2.02	77.33 ± 1.48	87.67 ± 1.12	70.17 ± 1.85
2	97.67 ± 2.14	111.83 ± 3.11	112.50 ± 1.61	110.17 ± 1.38	98.50 ± 1.93
3	105.50 ± 2.67	121.33 ± 2.50	120.50 ± 2.38	123.83 ± 1.66	106.83 ± 2.64
4	111.00 ± 2.58	121.33 ± 2.17	119.50 ± 1.41	122.33 ± 1.65	111.83 ± 1.52

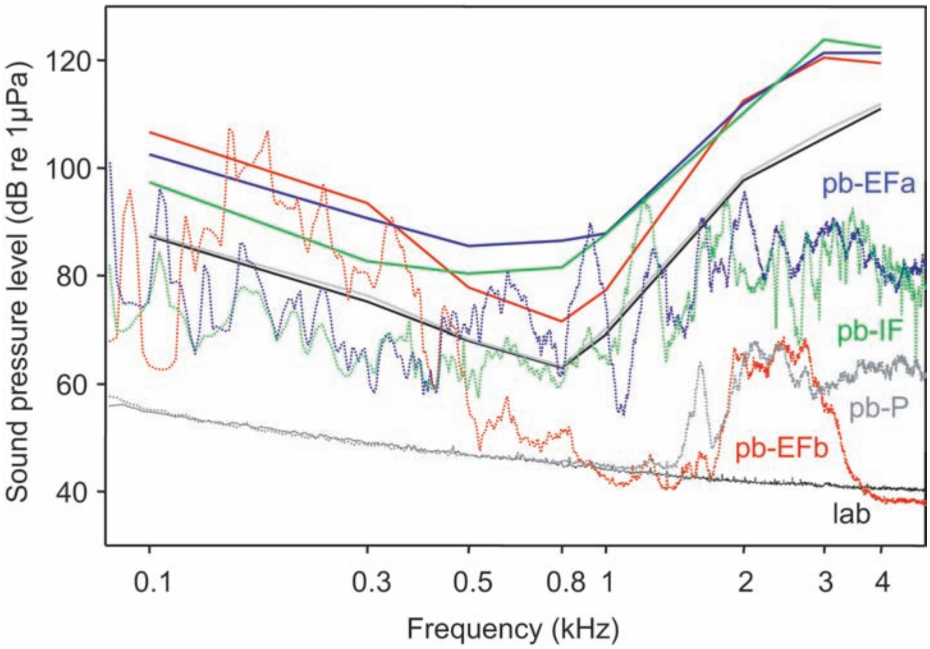

Figure 4. Mean hearing thresholds of *Carassius auratus* under laboratory conditions (baseline) and in the presence of the different artificial noise types (solid lines) and the sound power spectra of the corresponding noise types played back during hearing thresholds determinations (broken lines). pb-EFa, playback of the noise from the external filter with outflow above water surface; pb-EFb, playback of the noise from the external filter with outflow below surface; pb-IF, playback of the noise from the internal filter; pb-P, playback of the noise from the pond.

surveys of underwater ambient noise measurements in aquaculture systems. Bart *et al.* (2001) found that mean broadband SPLs differed across various intensive aquaculture systems. These levels varied from <100 dB re 1 µPa in an earthen pond with the aerator turned off, 120 dB re 1 µPa RMS in concrete raceways, to 130 dB re 1 µPa in round fibreglass tanks of various sizes. They observed the highest noise levels in intensive recirculation culture systems with large (14 m-diameter × 4 m deep) fibreglass tanks (153 dB), and in a pond system with the aerator turned on (135 dB). Electric paddle wheel aerators contributed significantly to the noise levels in the outdoor ponds.

Noise in private aquaria where ornamental fish are kept have not been investigated so far and only one study has investigated noise in public aquaria according to our knowledge. Anderson (2009) surveyed the noise conditions in nine public aquaria in the USA where tank sizes, aquaria wall materials, bottom type, filtration and aeration varied widely and found that noise levels ranged from 116 to 143 dB re 1µPa when hydrophones were positioned in the middle of the aquaria.

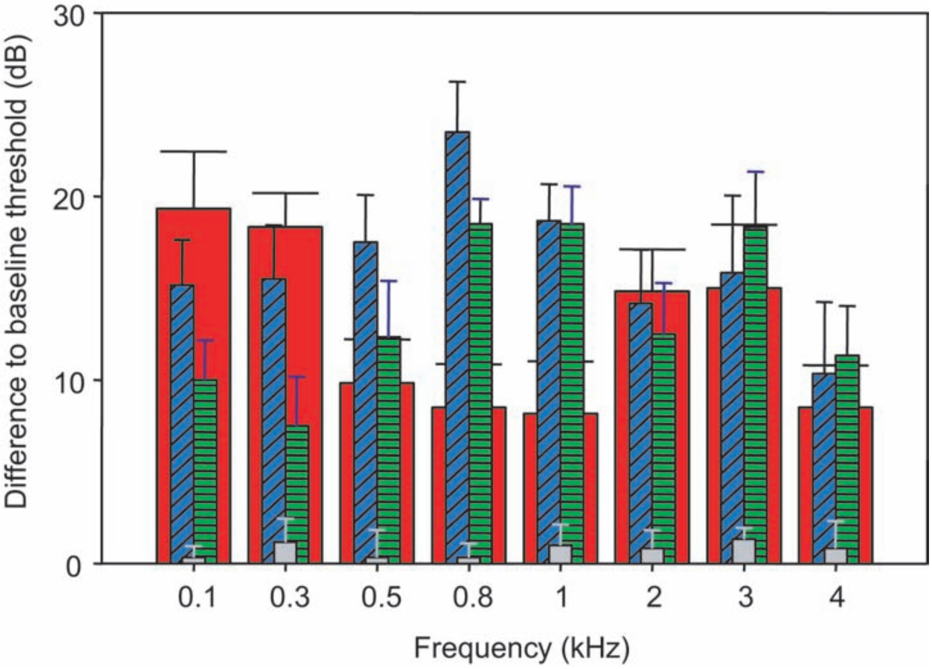

Figure 5. Differences in hearing thresholds between the baseline audiogram and the masked audiograms. Values are means ± S.E.M. (*N*=6). Bar types indicate the differences for the respective noise type according to Fig. 3. Broad bar in background = EFb (external filter with outflow below surface); oblique striped bar = EFa (external filter with outflow above water surface); horizontally striped bar = IF (internal filter); plain narrow bar in foreground = P (pond).

The noise levels encountered in different filter setups in the present study varied between 113.4 to 119 dB and thus are generally lower than in most public aquaria. The setup of the filtration system influenced SPLs. The noise level created by the internal filter with the outflow below the water surface increased when the aeration rate was maximized (114-119 dB). The external filter with the terminally plugged, above-water outflow (water exited the pipe only via lateral holes) was louder than with the underwater outflow. The SPL of the plugged outflow was higher than without a plug because numerous small water jets are louder than a large single outflow. This means that the SPL will always be higher if more air gets into the water.

Furthermore, higher frequency and more complex spectral components were observed in the aquaria with different filtering conditions than in the pond. Spectral levels of the EFa setup were higher above 0.5 kHz than those of EFb and IF setup because of more oscillating and collapsing air bubbles.

There was a maximum difference in noise levels of more than 40 dB between the spectral levels of EFa and the others (maximum difference between EFa and P ($\Delta_{EFa\text{-}P}$): 46 dB at a frequency of 0.15 kHz; $\Delta_{EFa\text{-}IF}$: 43 dB at 1.7 kHz; $\Delta_{EFa\text{-}EFb}$: 41 dB at 1.6 kHz). The maximum difference of the spectral noise levels in a concrete and fibreglass tank was about 15 dB at 250 Hz (Terhune *et al.* 1990), whereas differences of up to 40 dB at 800 Hz were found in public aquaria (Anderson 2009).

Low-frequency noise is generated by water flows, ground vibrations, aquarium wall vibrations and electrical pumps and filter motors (Bart *et al.* 2001; Davidson *et al.* 2007). Lower-frequency sound, below 0.1 kHz, with spectral levels of 74-110 dB in the aquaria and 60-83 dB in the pond were detected. Bart *et al.* (2001) measured SPLs of 125-135 dB re 1 μPa below 400 Hz.

In summary, minimizing the aeration of an internal filter with the outflow below the water surface decreased the SPL. In order to lower noise levels in aquaria, the following measures could be taken: (1) the water outflow pipe of the external filter should be close to or below the water surface and not plugged. (2) If the outflow is above the surface, it should not be vertically steered into the water (Table 1). (3) External filters should have no contact to the table on which the aquarium is standing. It proved advantageous to place the external filter on the floor (Table 2). Thus, using a quiet filter setup can help to reduce noise levels in the water.

Hearing under noise conditions

Hearing in our study animals was affected differently by the four noise conditions. The goldfish did not show masking effects during the presentation of pond noise. Noise levels in the pond were low (L_{Leq} < 100 dB) because it lacked an aerator or filtering system. Ponds represent seminatural habitats in which goldfish and other cyprinids such as koi carps are often kept for leisure all over the world. The good hearing capabilities of otophysans are well adapted to quiet habitats, and these fish can detect low-level sound produced by prey or food items and by con- or heterospecifics (Amoser & Ladich 2005; Wysocki & Ladich 2005a).

Contrary to the pond, aquarium noise had pronounced effects on auditory sensitivity. Hearing in goldfish was heavily masked under all aquarium noise conditions. Hearing thresholds at every measured frequency were masked by at least 8 dB. In the low-frequency range (0.1 and 0.3 kHz), thresholds were highly masked because of the abundant low-frequency noise. In the best hearing range of goldfish (0.5, 0.8 and 1 kHz), the amount of threshold shift decreased when a quieter water outflow, e.g. below the surface, was used; spectral analyses also showed a reduced spectral noise level in this frequency range.

The extent of threshold shift increased for EFa and IF noises because of the higher spectral noise levels caused by the stronger aeration, i.e. by oscillating and collapsing air bubbles. Above 1 kHz, all hearing thresholds were masked by about 13 dB because of the strong high-frequency underwater noise generated by splashing water noise and by oscillating and collapsing air bubbles. As indicated by the two-way ANOVA, the various types presented had different masking effects at the various frequencies tested. This can be explained by spectral differences among noises.

Hearing specialists are masked to a larger extent; they cannot exploit their excellent hearing abilities in environments with high SPLs (Popper & Fay 1993; Scholik & Yan 2001; Ladich & Popper 2004; Amoser & Ladich 2005, 2010; Wysocki *et al.* 2005a; Scholz & Ladich 2006). The results of the current study indicate that hearing specialists such as the goldfish are considerably masked under artificial holding conditions either in private aquaria or in aquaculture facilities. Wysocki *et al.* (2006) showed that cyprinids are susceptible to noise-induced stress response.

Species lacking accessory hearing structures (air-filled cavities connected to the inner ear) to enhance auditory abilities essentially respond to low-frequency sounds (and only below 1 kHz) at relatively high sound intensities (Hawkins & Myrberg 1983; Ladich & Popper 2004; Popper & Fay, in press). They exhibit their best hearing range at lower frequencies than specialists, and have worse hearing thresholds throughout the audiogram. According to prior findings (Amoser & Ladich 2005; Wysocki & Ladich 2005a), non-specialists would be only moderately or not at all masked in the presence of different aquarium noise types found in the present study. Wysocki & Ladich (2005a) showed that masking was low in the presence of white noise. The European Perch *Perca fluviatilis*, a non-specialist, was barely masked by quite different ambient noise types in any aquatic habitat due to its low hearing sensitivity (Amoser & Ladich 2005). Similarly, tank noise had no influence on the development of hearing thresholds in Rainbow Trout *Oncorhynchus mykiss* reared in 115 dB versus 150 dB tanks (Wysocki *et al.* 2007b). Despite the lower hearing sensitivities of species lacking accessory hearing structures, Wysocki *et al.* (2006) and Anderson (2009) observed physiological stress response in percids and syngnathids under noisy conditions.

Anthropogenic noise not only masks hearing under artificial holding conditions in aquaria and aquaculture facilities, it is also increasing in natural habitats of fishes and affects different fish species. Noise emanating from ships masks hearing in a number of non-related fish groups in several coastal regions. Low-frequency ship noise masks hearing and therefore the detection of conspecific sounds in representatives of Sciaenidae (*Sciaena umbra*) and Pomacentridae (*Chromis chromis*) in the Adriatic Sea (Codarin *et al.* 2009) and of Batrachoididae (*Halobatrachus didactylus*) in the coastal regions near Lisbon (Vasconcelos *et al.* 2007). These two studies are the first indication that anthropogenic noise also impacts acoustic communication in marine fish families inhabiting the European coast. These and our results show that anthropogenic noise apparently impairs fish hearing in all aquatic environments.

CONCLUSION

Fishes kept for leisure are exposed to different levels and spectra of background noise. We observed higher and more complex noise spectra in filtered aquaria than in the pond. Low-frequency underwater noise was generated mostly by the motors of the filters, whereas high-frequency noise was probably due to oscillating and collapsing air bubbles (Bart *et al.*, 2001).

Current data show that goldfish are heavily masked under artificial holding conditions and cannot exploit their excellent hearing abilities in environments with high noise levels. Aerators and other sound sources in aquaculture systems can be configured in a way that only minimally affects fish physiology (masking, stress) and growth. Decreasing the SPLs requires minimizing the splashing of water, positioning the outflow pipe close to or below the water surface, and not plugging the end of the pipe. These arrangements considerably lowers masking in the best hearing range of the goldfish (0.5-1 kHz). The filter should not be in contact with the aquarium table. It is advisable to place the filter below the aquarium on styrofoam or on other soft material rather than positioning it directly next to the aquarium.

ACKNOWLEDGEMENTS

We want to thank Heinz Tunner for providing goldfish and enabling us to measure and record ambient noise in his pond, Sonja Amoser for her help with sound recordings and Michael Stachowitsch for scientific English proof reading. This study was supported by the Austrian Science Fund (FWF grants No. P 17263 and 22319 to F.L.).

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Received 23 March 2010, revised 30 June 2010 and accepted 9 July 2010