were transferred to a holding chamber where a 0.015% solution of the anesthetic was passed through the mouth and over the gills of the fish. The caudal portion of the medulla oblongata and rostral spinal cord were exposed, and a microelectrode filled with Lucifer yellow (Sigma; 5% in distilled water) was inserted into the soma of only one neuron. Since the neurons exposed in this area of the central nervous system could either be in a supramedullary position or a spinal cord position, we will refer to the cells as supramedullary/dorsal neurons. The dye was iontophoresed (−10 nA of current was pulsed for 200 ms at a rate of 3/s) for about 1 h. About 1 h after injection the fish were perfused with 10% phosphate-buffered formalin. The brain and spinal cord were then dissected out, dehydrated, and cleared with methyl salicylate. The whole brain was viewed with a fluorescent microscope.

The somata of supramedullary/dorsal cells are visible on the surface of the brain with the aid of a dissecting microscope. In seven fish, a single soma was located and filled with dye. Lucifer yellow traveled from the filled cell to adjacent ones in three of the seven fish. One cell rostral and two cells caudal to the filled cell contained dye in two fish. In the third fish two cells rostral to the filled cell contained dye (Fig. 1). In the four cases where there was no apparent dye transfer between neurons, the intensity of the fill appeared similar to that of filled neurons in which dye coupling did occur. To control for the possibility that extracellular leakage of the dye might label neurons other than the one being filled, a single dorsal gill was penetrated with a dye-filled microelectrode in two fish. The electrode was withdrawn to just outside the cell membrane and Lucifer yellow was iontophoresed extracellularly for 1 h, the fish was perfused, and the brain processed as described above. No dye was localized to any cell.

The transfer of Lucifer yellow from one supramedullary/dorsal cell to others provides morphological evidence for the existence of gap junctions. The lack of dye coupling in four fish does not necessarily mean that gap junctions do not exist between supramedullary/dorsal cells in these fish. For example, there may be a wide distribution of sites of electrical coupling, or Lucifer yellow may not have crossed the gap junctions (3, 4). When dye coupling occurred, the fall-off in dye concentration from the filled cell to adjacent neurons was large, so more distal neurons may be equally well coupled but not contain dye. The cunner has between 35 and 40 supramedullary/dorsal cells. Electrical coupling measurements will help determine the extent of coupling between this group of neurons. Supramedullary/dorsal cells in the cunner are sensitive to tactile stimulation (5). Our results predict that neurons that are electrotonically coupled will fire synchronously with sufficient tactile stimulation.

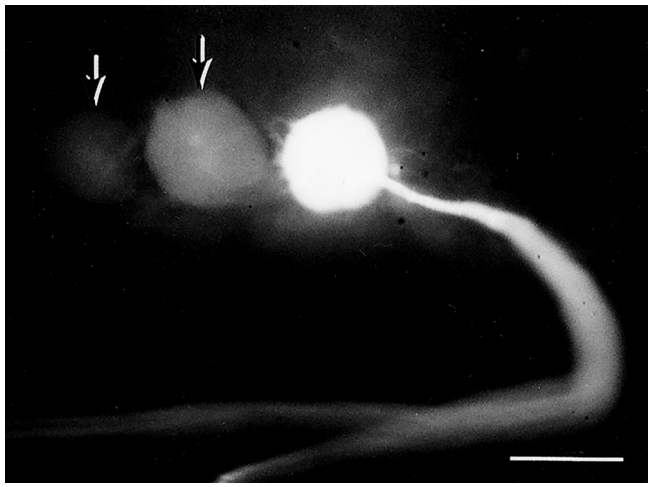

Figure 1. *Lucifer yellow injection of a single supramedullary/dorsal cell in the cunner. The cell on the far right was iontophoretically filled with dye; after fixation, dehydration, and clearing, the whole brain was viewed with a fluorescent microscope. The soma of the filled neuron gives rise to a single process that extends ventrally and bifurcates near the bottom of the photomicrograph. Two other rostral somata (arrows) contain dye as well, providing support for the existence of gap junctions between these cells. This is a sagittal view of the brain with dorsal up and rostral to the left. Calibration bar = 100 μm.*

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A Comparison of Sounds Recorded From a Catfish (*Orinocodoras eigenmanni,* Doradidae) in an Aquarium and in the Field

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Parvulescu (1) raised concerns regarding the suitability of a small glass aquarium for characterizing fish sounds based upon a theoretical consideration of sound echoes. Four out of eight authors who cited this paper most recently noted that small aquaria have complex acoustics, and the other four described the aquarium environment as yielding imprecise and poor quality sound recordings. Further advances in the study of sound production and communication in fishes require studies in controlled laboratory environments. Recently, Lugli (2) noted that waveforms and sound spectra were similar for field- and aquarium-recorded goby sounds.

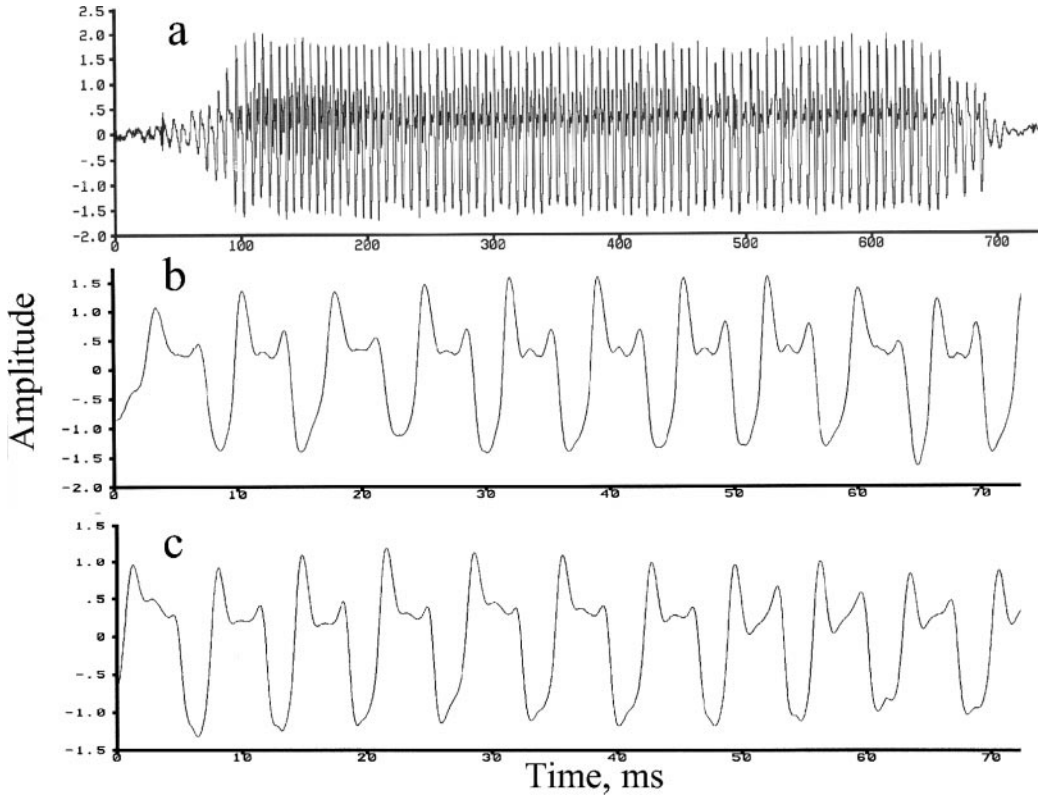

Figure 1. *Swimbladder disturbance sounds for three different individuals of a doradid catfish,* Orinocodoras eigenmanni: *(a) waveform of one entire sound, field recording, (b) expanded waveform of 10 pulses, field recording (c) expanded waveform of 10 pulses, aquarium recording. The time scale differs between the top (a) and bottom two plots (b & c) by a factor of 10.*

Okumura *et al.* (3) observed that artificially generated sounds recorded close to a hydrophone were free from acoustic artifacts. Whether more complex, natural fish sounds would also be artifact-free requires testing. We elicited sounds from a catfish and compared field and aquarium recordings which were specifically made close to the signal source.

We analyzed sounds produced by the swimbladder mechanism of a catfish in the disturbance context (fish are restrained by a human hand) underwater. Similar sounds were produced by the same fish in conflicts over resting sites (4). Many fishes that produce sounds in intraspecific behavioral contexts also "release" these sounds when restrained (5). We chose swimbladder sounds because they are a common mechanism of sound production for many fishes (6, 7).

We recorded sounds of nine individuals of a wild-caught neotropical catfish, the doradid *Orinocodoras eigenmanni.* Standard length ranged from 5.7 to 8.5 cm. Each individually recognizable fish was recorded twice in both recording environments. Recordings were conducted during 10 July–6 August 1992. Fish were positioned 7.5 cm from a hydrophone and 23 cm under the water surface. Fish were held with their left side toward and their swimbladder centered on the midpoint of the hydrophone. In the field (Jenkins Pond, Falmouth, MA) fish were recorded in a containment net. The net had a 60-cm diameter and 60-cm maximum depth. Water depth at the dock field site (Jenkins Pond) was 90 cm over a sand bottom. Aquarium recordings were conducted in a 10-gallon glass aquarium on a grass lawn near the pond. The hydrophone was suspended in the center of the water-filled aquarium. Fish were held in the same relative position to the hydrophone and water surface as in the field. Temperatures for recording dates in the aquarium and in the field were not different (24.7 ± 0.6 aquarium, 25.2 ± 0.3 field; $n = 3$). Sounds were recorded using a tape recorder (SONY Model WM-D6C: frequency response 40–15,000 Hz $\pm$ 3 dB). The hydrophone was pressure sensitive and had a frequency response range of 10 to 3,000 Hz (BioAcoustics, see 8 for specifications). The acoustic analysis software SIGNAL (Engineering Systems, Belmont, MA) was used to digitize and analyze sounds (sampling rate 25 kHz). We only analyzed sounds which had clear pulse structure. Both recording environments occasionally yielded some sounds with obscured pulse number and waveform patterns, due to spurious background noise or fish movements.

Spectrograms of over 800 sounds were evaluated (580 field, 275 aquarium). The catfish produced similar numbers of sounds in both recording environments. A minimum of ten sounds were produced by each individual on each sampling date. The same types of sounds were produced by individuals in both recording environments. Sound duration ranged from 30 ms to 2,400 ms.

In order to assess whether sounds were altered in the aquarium environment compared to the field, we compared waveforms visually and pulse durations statistically for sounds from both recording environments. Waveforms of sound pulses for field and

aquarium showed the same shapes (Fig. 1). No artifacts were noted. Pulse duration was measured for one sound per individual ($n = 9$) for seven pulses in the center third of the sound where pulse peak amplitudes were consistent. Individual pulse durations ranged from 6 to 7 ms and were not significantly different between field and aquarium environments (one way ANOVA). For aquarium-recorded sounds, the pulse duration mean was 6.5 (SE 0.07, $n = 63$). For field-recorded sounds, the pulse duration mean was 6.5 (SE 0.07, $n = 63$).

Disturbance context swimbladder sounds of a catfish showed no differences in pulse waveform or pulse duration when recorded close to a hydrophone in both field and small aquarium recording environments. Kastberger (9) observed that for field recordings of doradid sounds, pulse pattern was unchanged for up to 30 cm. Many fishes initiate sound production in close proximity to conspecifics (10, 11). These results suggest that a small aquarium environment can provide sound recordings that accurately represent the sounds a fish produces in the field, yielding reliable acoustic measurements.

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Bimodal Units in the Torus Semicircularis of the Toadfish (*Opsanus tau*)

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We have been investigating aspects of auditory processing and directional hearing in the toadfish *Opsanus tau.* We have shown that the saccule is an auditory endorgan that encodes both frequency and direction of a sound source (1). This information is sent *via* the VIIIth nerve to nuclei in the medulla, in particular, the descending octaval nucleus (1). Our previous work on cells in the descending octaval nucleus in *Opsanus tau* has revealed that most are highly directional (1) and that these directional auditory cells project to the midbrain. The torus semicircularis (TS) is a sensory processing site in the midbrain of fishes and amphibians. Nucleus centralis in the TS receives input from auditory areas in the medulla, and nucleus ventrolateralis receives input from lateral line areas in the medulla (2). Here we report some preliminary results from extracellular recordings of auditory cells in the TS.

Our protocol is described in detail elsewhere (1). In brief, the toadfish is anesthetized and immobilized (pancuronium bromide injection and lidocaine applied topically), and the dorsal surface of the midbrain is exposed. Following surgery, the fish is placed in a cylindrical dish filled with fresh seawater and is secured with a head holder. The water surface in the dish lies just below the surgical opening in the skull. The dish is part of a three-dimensional shaker table that provides sinusoidal motion of the animal with the surrounding water along linear pathways to simulate the particle motion component of underwater sound at appropriate frequencies (50–300 Hz) and levels, in the horizontal and midsagittal planes at specified angles (0°, 30°, 60°, 90°, 120°, 150° in each plane). In addition, we tested for external mechanoreceptive sensitivity (tentatively identified as lateral line) by producing hydrodynamic disturbances using puffs of air at the water surface along the length of the fish in the absence of an auditory stimulus. Units were classified as responding to hydrodynamic stimuli if the evoked spike rate was two standard deviations or more above the mean background rate.

For extracellular recording we used pulled glass electrodes with tip sizes of 3–5 μm and resistances of 3–10 MΩ. Our recording sites in the TS were confirmed in two ways. First, we used neurobiotin-filled electrodes (4% in 3 *M* NaCl) to mark the location of the first auditory cell analyzed. Second, the location of the electrode at all recording sites was plotted using the scale on a three-dimensional micromanipulator (accuracy to 10 μm). The neurobiotin was visualized using standard ABC immunohistochemistry (Vector Labs) in 50-μm floating sections, which were then placed on slides, dehydrated, and coverslipped.

We have recorded from 71 units in the TS. Of the cells that responded to the auditory stimuli, we have found that 33% have auditory sensitivity only and 67% respond to both auditory and hydrodynamic stimulation. Units unresponsive to auditory stimuli but responsive to hydrodynamic stimuli were observed frequently, but were not analyzed further. Figure 1 illustrates the responses of two TS units to varying levels of whole-body vibration in three orthogonal directions and to the hydrodynamic assay for putative lateral line sensitivity. Some units demonstrate a relatively large