

Sound source localization by the plainfin midshipman fish, *Porichthys notatus*

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The aim of this study was to use plainfin midshipman fish (*Porichthys notatus*) as a general model to explore how fishes localize an underwater sound source in the relatively simple geometry of a monopole sound field. The robust phonotactic responses displayed by gravid females toward a monopole sound projector (J-9) broadcasting a low-frequency (90 Hz) tone similar to the fundamental frequency of the male's advertisement call were examined. The projector's sound field was mapped at 5 cm resolution azimuth using an eight-hydrophone array. Acoustic pressure was measured with the array and acoustic particle motion was calculated from pressure gradients between hydrophones. The response pathways of the fish were analyzed from video recordings and compared to the sound field. Gravid females at initial release were directed toward the sound source, and the majority (73%) swam to the playback projector with straight to slightly curved tracks in the direction of the source and in line with local particle motion vectors. In contrast, the initial direction of the control (sound-off) group did not differ from random. This paper reports on a comparison of fish localization behavior with directional cues available in the form of local particle motion vectors. © 2010 Acoustical Society of America. [DOI: 10.1121/1.3365261]

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I. INTRODUCTION

Fish, like most animals, live in highly complex acoustic environments that require them to make sense of the objects and events that take place there. In order to behave appropriately toward the objects and events, animals must be able to “segregate” streams of acoustic information so that independent sources are not confused and that the “scene” is understood. An important component of stream segregation is sound source localization. It is the ability to localize sound sources that gives the acoustic scene a spatial dimension. Evidence suggests that the capacity for sound source localization is common to mammals, anurans, birds, and reptiles, but surprisingly it is not known whether fishes locate sound sources in the same manner. The focus of the present study was to investigate sound source localization by fishes, and as a first step explore how fish localize an underwater sound source in the relatively simple geometry of a monopole sound field.

The question of sound source localization by fishes was first raised in 1935 by Reinhardt (1935) and by von Frisch and Dijkgraaf (1935). Both groups worked with the European minnow (*Phoxinus laevis*), and both concluded that *Phoxinus* could not localize sound sources. The European minnow is a pressure-sensitive otophysan, having Weberian ossicles that connect the swim bladder to the inner ears (Weber, 1820). Otophysans were thought to respond exclusively

to sound pressure as transduced by the swim bladder and transmitted as motions to the ears via the Weberian ossicles, and von Frisch (1938) explained the negative result of the behavioral experiments by pointing out that sound pressure at any single point (such as the swim bladder) contains no information about the direction of sound propagation. Other modes of acoustic stimulation of fish inner ears were not known or understood at the time; thus, sound pressure sensitivity was considered an essential part of the definition of hearing; a definition that continued to dominate for some time under the influence of van Bergeijk (1964).

Presently, hearing in fishes is somewhat better understood, and it is generally accepted that fishes have at least two modes of hearing. One mode is pressure-mediated and found either in species with specialized skeletal adaptations (e.g., the Weberian ossicles) that connect the swim bladder to the inner ear or in species where the swim bladder, or other gas-filled structure(s), is in close or direct contact with the ear. The other mode of hearing is ancestral (shared by all fishes) and based on the inertia of otolith organs (De Vries, 1950). The inertial mode of hearing is independent of the swim bladder. It results from direct stimulation of the inner ear by acoustic particle motion in the near and far fields (e.g., Popper and Fay, 1993). During inertial-mode stimulation the entire fish is set into motion by the acoustic disturbance but impedance differences between the otoliths and surrounding

tissue cause relative movement between the otolith and the bed of hair cells in the sensory epithelium (macula). The auditory hair cells are inherently directionally sensitive and are organized in patterns where the axis of best directional sensitivity varies along the maculae of the end organ. Thus, it is this inertial mode of hearing that could presumably provide directional information to fish (Edds-Walton *et al.*, 1999).

The behavioral capacity for directional hearing by fishes has been demonstrated in several species using psychoacoustic discrimination techniques. Direction-dependent masking was shown (Chapman, 1973; Chapman and Johnstone, 1974), minimum audible angles of 15°–20° in both azimuth and elevation were measured (Schuijf, 1975; Hawkins and Sand, 1977), and source distance discrimination was demonstrated (Schuijf and Hawkins, 1983). If it is assumed that sound source localization by fish is essentially similar to that in humans, in the sense that observers “know” where the sources to be discriminated are located and can act on this information, then the conditioned discrimination experiments provide evidence for good localization ability that is probably widely shared among fishes. However, Kalmijn (1997) argues for source localization strategies by fishes that do not assume familiar, human-like capacities and behaviors, but instead are based on ethological tenets and what is presently accepted about source localization using the ampullary electro-sensory system. Kalmijn points out that fish could find their way to a sound source by swimming in a direction that maintains a constant angle between the fish and the axis of particle motion. The fish does not have to “know” the source location or the orientation of the local particle motion vector, it just has to behave in a way that maintains a constant angle with the particle motion vector and it will eventually arrive at the source. The question of whether fish “know” where a source is located or use a more mechanistic approach remains open, in part, because few experiments have demonstrated that fish can use sound to move to a source and no experiment has compared the pathways fish take to a sound source with the directional cues available in the particle motion vector field.

McKibben and Bass (1998, 2001) found that gravid females of the species *Porichthys notatus* exhibit robust phonotaxis toward an underwater loudspeaker broadcasting a male advertisement call (a continuous “hum” consisting of a multiharmonic signal with a fundamental frequency of about 90 Hz), or a low-frequency tone near the fundamental frequency of the male’s advertisement call. Gravid females often respond to the tone by approaching the sound source and either directly touching the speaker face or by circling in front of or underneath the playback speaker. This unambiguous phonotactic response by gravid females makes the midshipman fish an exceptional model animal for investigating sound source localization. Also, because these fish will perform in a controlled (laboratory) setting, the sound fields can be mapped, including the particle motion vectors, to produce a comprehensive map of the acoustic environment.

The primary objective of this study was to investigate how female plainfin midshipman fish (*P. notatus*) localize a sound source in the relatively simple sound-field geometry

produced by a monopole source. The phonotactic responses of reproductive females are described in relation to the particle motion and pressure components of the measured sound field. The pathways that the fish take to the source could indicate their near-field decision parameters and sound source localization strategies.

II. MATERIALS AND METHODS

A. Experimental animals

Female plainfin midshipman fish were collected during the reproductive season (June and July in 2007 and 2008) from the same geographical locations used in previous studies along Tomales Bay near Marshall, CA (Brantley and Bass, 1994; McKibben and Bass, 1998; Sisneros and Bass, 2005). Females were collected by hand from the nests of type I males in the intertidal zone at low tide and were readily distinguished from nesting type I and sneaker type II males based on the size of the animal and shape of the urogenital papilla. Females collected from nests were in various stages of spawning, but the majority of the collected females were gravid (full of eggs) with ovaries that contained relatively large (~5 mm in diameter) yellow, yolked eggs. Gravid females were also visually distinguished from type I males based on the size and appearance of the abdomen, which were typically distended in gravid females and noticeably flaccid in spent females (Brantley and Bass, 1994; Bass, 1996). The reproductive state of females was quantified by measuring the gonadosomatic index (GSI, defined here as 100* gonad mass/body mass—gonad mass, according to Tomkins and Simmons, 2002). Animals collected in the field were temporarily housed (for a few hours) in coolers with aerated seawater until they could be brought to the Bodega Marine Laboratory in Bodega Bay, CA, where they were maintained in large communal aquaria at natural ambient temperatures that ranged from 12 °C to 14 °C. Fish were held for less than 20 h prior to testing later that night.

B. Experimental tank and setup

All tests were conducted outdoors in a cylindrical concrete tank (4 m in diameter, 0.75 m in depth) at the Bodega Marine Laboratory, Bodega Bay, CA. A monopole sound projector (U.S. Navy J-9 transducer) was suspended from a beam in the center of the tank with the center of the projector face positioned 7 cm above the tank floor and facing outward (Fig. 1). A 2.44 m opaque plastic tarp was used as a screen and placed immediately in front of, but not touching, the sound projector. This was done to remove any visual cues that might affect sound source localization behavior.

The playback signal consisted of a continuous tone at 90 Hz that was similar to the fundamental frequency of the male advertisement call (80–100 Hz; Bass *et al.*, 1999). The playback signal of 90 Hz was chosen for this study based on the finding that there is a temperature-dependent frequency preference for the phonotactic response (McKibben and Bass 1998). The acoustic stimuli for the playback experiments were generated by a Wavetek function generator and passed through a power amplifier (Crown Audio, Inc., Elkhart, IN)

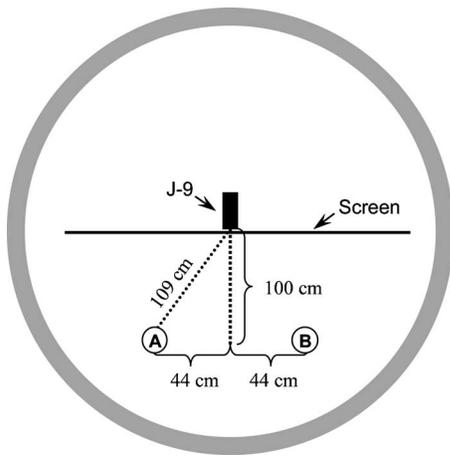


FIG. 1. Schematic of the playback tank (diameter=4 m; tank depth =0.75 m) showing the sound projector (J-9) position, opaque screen, and animal release sites (A and B).

that drove the U.S. Navy J-9 sound projector. The sound level of the J-9 projector was monitored and nightly calibrated using a custom-written LABVIEW program with a National Instruments 6052E DAQ-Pad (Austin, TX) and a mini-hydrophone (8103, Bruel & Kjaer, Norcross, GA) placed 4.5 cm above the tank floor on the center line between the two animal release sites (Fig. 1). The tone level at the calibration site was set at 130 dB (re 1 μ Pa peak) and is consistent with sound pressure levels of the advertisement calls of type I males recorded near their nests (Bass and Clark, 2003).

The behavioral responses of the fish were recorded on videotape using a video recorder and a CV110 Precision black-and-white camera (0.2 lux \times minimum light level) mounted approximately 6 m above the tank's test arena. A remote positioning system was used to direct and position the camera above the test arena. The video records were digitized with a Vixia HV30 camcorder (Canon Inc., Lake Success, NY) and IMOVIE 7.0 software (Apple Inc., Cupertino, CA). Windows MOVIE MAKER 5.0 (Microsoft, Redmond, WA) and SIGMASCAN PRO 5.0 (Systat Inc., Chicago, IL) were used for frame-by-frame analysis of the digitized video records. Every fifth frame was analyzed by marking the position of the animal's head (on the midline between the two eyes) relative to the fixed position of the sound projector. The x - and y -coordinates of the animal's head were then used to track the movement of the animal (behavioral response) in relation to the measured sound field.

C. Experimental protocol

The sound playback experiments were conducted at night between 21:00 and 2:00 h during the midshipman summer breeding season from June through July. Three red floodlights positioned around the tank perimeter allowed for the observation and videotaping of the behavioral responses from female midshipman fish. The water temperature in the test tank was controlled by adjusting the incoming flow rate of seawater to the tank prior to the behavioral tests. The water flow to the test tank was shut off during all tests. Water depth was adjusted to 50 cm for all tests.

Prior to testing, female fish were held individually in 5 gallon buckets with water from the test tank at the test tank temperature and were allowed to acclimate for at least 10–15 min. Tests began with an individual fish being placed in a 30 cm in diameter plastic mesh cylinder positioned approximately 109 cm from the sound source (Fig. 1) while the sound playback stimulus was already playing. Fish were then released by manually raising the cylinder. This protocol of releasing the fish without an acclimation period while the acoustic stimulus was continuously playing was adopted from McKibben and Bass (1998). Tests were terminated when the fish swam to the perimeter of the testing arena or when the sound was turned off after a positive phonotactic response. A positive response was recorded when a fish approached the sound source and then directly touched the speaker face or circled in front of or under the sound projector. There were no observations of fish returning to the center of the tank after reaching the periphery, and rarely, if ever, did a fish remain in the center of the tank when the speaker was off.

D. Acoustic and vibration measurements

Pressure measurements were made with miniature (50 mm in length, 9.5 mm in diameter) hydrophones; either Bruel & Kjaer type 8103 or Reson type TC4013. These hydrophone types are essentially identical, both have a useful frequency range of 1–170 kHz and sensitivity of approximately -212 dB re 1 V/ μ Pa. The output signal from the hydrophones was amplified by two 4 channel charge amplifiers (Bruel & Kjaer model 2692) and fed into a DAQ pad 6052e (National Instruments). Custom LABVIEW (National Instruments) programs were written to control and record from the DAQ pad (program author D. G. Zeddies). The pressure calibration of each hydrophone was tested with a Bruel & Kjaer 4229 pistonphone calibrator. All hydrophones measured within 0.25 dB of the expected pressure and 0.25 dB of each other.

In order to determine substrate vibration introduced into the behavioral arena through mechanical coupling of the sound projector, vibrational amplitudes were measured using Geospace CT32 geophones (Houston, TX) that had a sensitivity of 0.197 V/cm/s. A VP2000 voltage preamplifier EC6081 (Reson, Goleta, CA) was used and the output signals recorded by a LABVIEW program via the DAQ pad.

1. Acoustic particle displacement measurements

The acceleration of the “particles” constituting the medium in which an acoustic disturbance takes place is proportional to the pressure gradient. This is formalized in the equation of motion, or Euler's equation (Pierce, 1994)

$$-\nabla p = \rho_0 \frac{\partial \mathbf{u}}{\partial t}, \quad (1)$$

where ∇ is the gradient operator on the pressure p , ρ_0 is the fluid density, \mathbf{u} is the velocity vector, and t is time. A discretized form of Euler's equation in one dimension is:

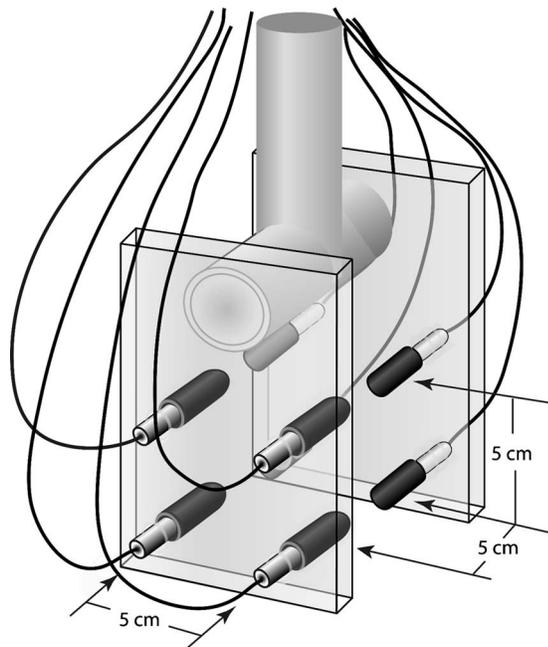


FIG. 2. Drawing of the probe used to measure sound pressure and determine pressure gradients within the behavioral arena. The probe holds eight hydrophones at separation distances of 5 cm in the x -, y -, and z -directions. The sides holding the hydrophones were constructed from plexiglass and were glued to PVC piping. The piping was attached to a cart that was used to move the probe around the tank. Plexiglass and PVC were chosen in an effort to keep the probe acoustically transparent at 90 Hz. Holes were drilled in the PVC piping (not show in drawing) to ensure that air was not trapped in the PVC piping that held the probe.

$$-\frac{P_1 - P_2}{\rho_0 d} = \frac{\partial \mathbf{u}}{\partial t} \quad (2)$$

where, P_1 and P_2 are the sound pressures measured at two points separated by a distance d , and \mathbf{u} is the velocity in line with the two points. Equation (2) lends itself to practical measurement because P_1 and P_2 can be measured with hydrophones. However, it should be pointed out that the complex pressure should be measured (magnitude and phase) and used in Eq. (2).

A probe was constructed using eight hydrophones (five Bruel & Kjaer type 8103 and three Reson type TC4013) to measure the pressure at known separation distances and directions. The hydrophones were arranged to form a 5 cm cube with a hydrophone at each corner (Fig. 2). This arrangement allows particle motion to be calculated in the x -, y -, and z -directions by finding the pressure gradient between adjacent hydrophones. A custom LABVIEW program was created to record the pressure and phase (relative to one hydrophone chosen as the reference) at each hydrophone, and then calculate the fluid particle displacement by integrating Eq. (2) with respect to time.

The phase angle measured for the pressure and used in Eq. (2) will depend, in part, on the response properties of the hydrophone. Differences in filtering among the hydrophones could bias the calculated particle motion. To determine the relative phase response of the eight hydrophones, a probe was made to hold two hydrophones along a radial line from the projector. The probe could then be rotated 180° so that

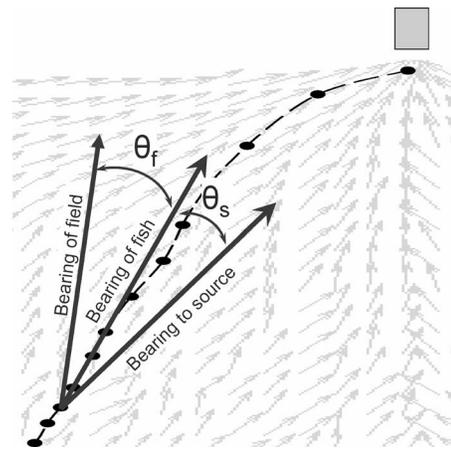


FIG. 3. An example showing how the difference angles of the fish's bearing relative to the sound source and to the local particle motion vectors were determined (see text for a more complete description).

the hydrophones exchanged places. Twenty 1 s samples with a 90 Hz tone (175 kHz sampling rate for each channel) were obtained. The standard deviation of the phase of each group in 20 trials was less than 0.25°. If the response time of the two hydrophones is the same, then rotating the probe 180° and adding the measured phases should equal zero. A deviation from zero would reveal the difference in response time between the two hydrophones and could be used as a correction factor. After rotating the probe 180° and taking another twenty samples, deviations from zero for the seven hydrophones measured relative to the reference were <0.5°. However, repeating the measurements revealed approximately up to 0.5° differences from prior measurements. The 0.5° deviations were more likely due to small placement errors when rotating the probe rather than actual differences in hydrophone response. For this reason no phase corrections were applied.

E. Analysis of movement

To analyze the movement of the fish, the difference angles of the bearing of the fish relative to the source and relative to the local sound field were determined (Fig. 3). This was done by finding the position of the fish from the video record (every fifth frame, 150 ms) and then calculating the fish's bearing between consecutive positional points. The difference angle relative to the source was the difference between the fish's bearing and the angle from the fish's position to the source. The difference angle relative to the local sound field was the difference between the fish's bearing and the bearing of the particle motion vector at the fish's position. These two angles were calculated between each time step (150 ms) for all recorded behavioral tracks.

As a measure of performance, the vector strength of the difference angles was computed. Vector strength is a measure of directional tendency or consistency toward the source (Batschelet, 1981); more formally, it is the normalized length of the mean vector of the circular distribution of angles to the source (the vector for each fish is unity length with the angle to the source, or in line with the particle motion vector). If all

directions are equally likely, the vector strength is zero, whereas if all fish move in the same direction, the vector strength is 1 (1.0).

III. RESULTS

A. Sound field measurements

To evaluate the phonotactic behavior of the fish, quantitative descriptions (maps) of the acoustic pressure and particle motion in the behavioral arena were obtained. Pressure is a scalar measure consisting of only a magnitude. It is the more familiar component because pressure is measured with a hydrophone. Particle motion (i.e., the displacement, velocity, and acceleration of the media due to an acoustic disturbance) is a vector, having both magnitude and direction. Particle motion must be determined with a vector sensor, such as an accelerometer, or can be calculated from the pressure gradient. In this study particle motion was calculated from the pressure gradient measurement using an array of hydrophones with known separation distances.

1. Acoustic pressure measurements

A continuous 90 Hz tone was used in all of the behavioral experiments. One-second samples with the tone on and with the tone off were recorded and the power spectral density calculated (using the supplied LABVIEW virtual instrument for power spectral density and sampled at 40 kHz). With the tone playing, the greatest spectral energy was found at 90 Hz (Fig. 4, top panel, arrow). The largest harmonic occurs near 270 Hz and is approximately 60 dB (1000 times) smaller than the primary tone at 90 Hz. Samples when no tone was played show 60-cycle noise and its harmonics, but they are small (Fig. 4, bottom panel).

To map the sound pressure field encountered by the fish during behavioral experiments, the pressure was measured at a height of 7 cm above the bottom of the tank at a resolution of 5 cm (a resolution of 22.5 cm was used behind the projector outside of the behavioral test arena). More than 1200 points were obtained in total. Figure 5 shows a contour plot of the sound pressure field for the standard “130 dB” level (at the release sites), the level at which the behavioral experiments were conducted. The sound pressure in the direct field near the projector is that expected for a monopole source in that the sound pressure level decays at the same rate in all directions as a function of radial distance. Farther from the projector, near the tank walls, the pressure becomes more variable, presumably due to reflections.

2. Acoustic particle motion measurements

Particle motion was calculated using the eight-hydrophone probe (the sampling rate was 175 kHz per hydrophone). With the eight hydrophones arranged in a cube, four measurements of particle displacement in each plane are obtained. Proper description of the particle motion is a vector in three-dimensional space. However, for ease of presentation, two two-dimensional space vectors were calculated in

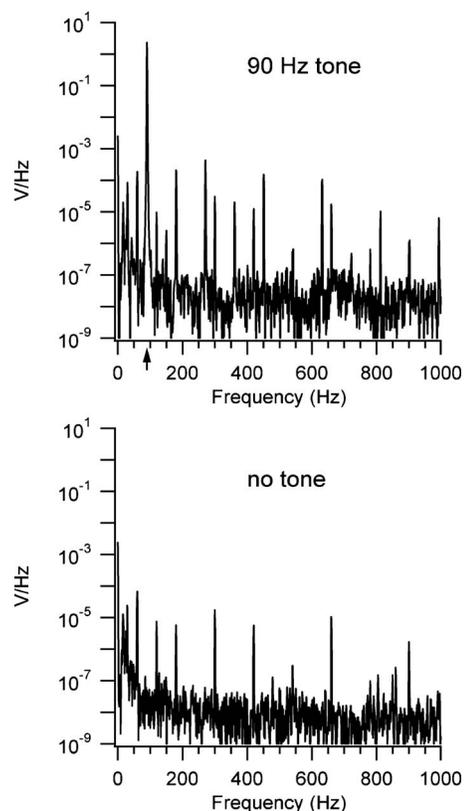


FIG. 4. Representative examples of the power spectral density from 1 s samples recorded 1 m from the source and centered between the release sites. The upper panel shows the power spectral density with the 90 Hz tone on, and the lower panel shows the power spectral density when the tone was off. Note that the greatest spectral energy was found at 90 Hz (arrow in upper panel) when the playback tone was present, and the background noise (lower panel) consisted of 60-cycle noise with harmonics that were approximately 60 dB or lower than the playback tone when present.

the XY plane (parallel to the bottom of the tank); one for the lower four hydrophones and one for the upper four hydrophones. The four z (normal to bottom of the tank) measurements were averaged into one magnitude.

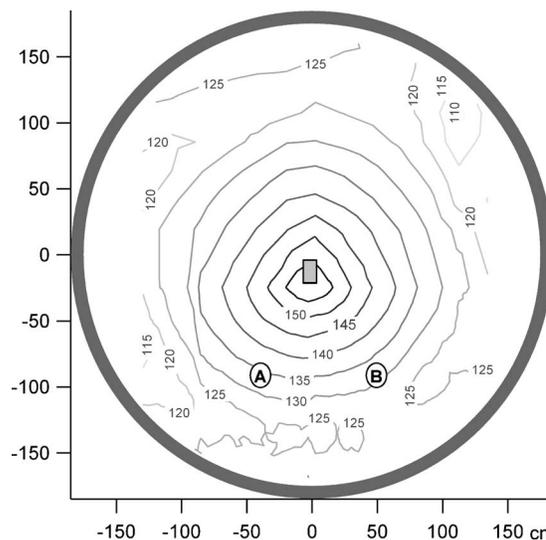


FIG. 5. Contour plot of the peak sound pressure level (SPL dB re 1 μ Pa) produced by the J-9 sound projector (■). The axes are the distance from the center of the tank in cm. Sound pressure was measured with the hydrophone array at 9.5 cm above the tank bottom. A and B are the animal release sites.

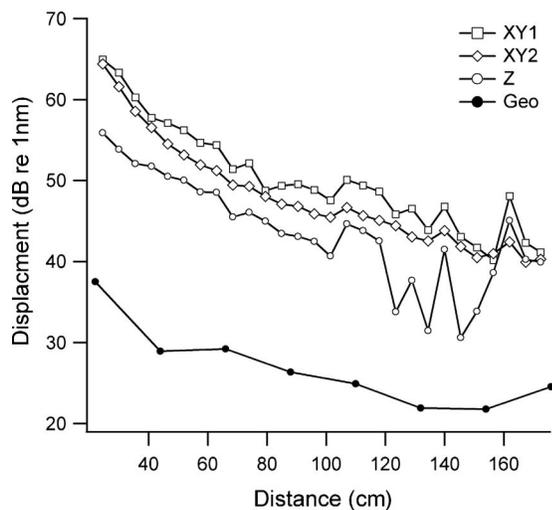


FIG. 6. Magnitude of the particle displacement (dB re 1 nm) along the radial between the release sites for the sound projector broadcasting a 90 Hz tone. XY1 and XY2 are the particle motion vectors measured at 9.5 and 4.5 cm above the tank floor, respectively. Z is the vertical particle motion, and Geo is the vertical substrate vibration measured with the geophone. Note that the substrate vibrations (Geo) are approximately 20 dB lower (10 times) than the particle displacement measured from the sound source at 9.5 cm (XY1) from the tank bottom.

Figure 6 shows the particle displacement magnitudes along the centerline of the tank in front of the projector. Particle displacement in the azimuth plane at 4.5 and 9.5 cm from the bottom of the tank are of similar magnitude and only a few decibels larger than the displacement magnitude in the *z*-direction. The substrate vibration displacement magnitude (measured using geophones) in the vertical direction for the tank bottom is approximately 20 dB less than the vertical acoustic displacement (Fig. 6). The vibrations on the beam suspending the projector, the tank wall, and at the release sites were 23.3, 28.5, and 21.9, respectively (A), and 23.2 (B) dB re 1 nm; all of which are less than the magnitude of the acoustic displacement in the vertical direction. The geophone measurements were done to determine if the vertical component of the sound field resulted from mechanical coupling of the projector to tank structures or as a result of the pressure release surface at the air-water boundary. Because the vertical motion measured with the geophone was less at all places than the vertical motion in the water column, it was concluded that sound introduced into the water from the projector was primarily responsible for vertical acoustic particle motion.

The vector fields of the *xy*-displacement magnitude and direction in the behavioral arena are shown in Fig. 7. It can be seen in Fig. 7 that, in general, the displacement vectors point toward the projector (or away: arrow heads were added arbitrarily to only one end of the drawing line) in the direct field. The magnitude of displacement was 65 dB re 1 nm 24 cm directly in front of the projector, and approximately 45 and 48 dB re 1 nm at the release sites A and B, respectively. It is also observed, that, presumably due to bottom interactions, the *xy*-displacement of the sound field measured at 4.5 cm from the bottom is more variable than the sound field measured at 9.5 cm.

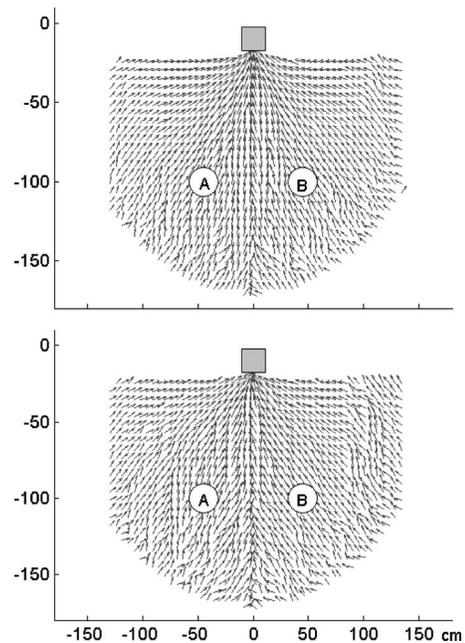


FIG. 7. Particle displacement vector field in the XY plane as measured at two distances above the tank floor. The axes are the distance from the center of the tank in cm. Upper panel shows the particle displacement measured at 9.5 cm above the tank floor. Lower panel shows the particle displacement measured at 4.5 cm above the tank floor. A and B are the animal release sites.

B. Phonotactic response pathways

Only gravid females ($n=45$, mean $GSI=22.9 \pm 8.0$ SD) exhibited positive phonotactic responses to the hum-like playback tone of 90 Hz. Spent females containing little or no eggs did not exhibit positive responses ($n=8$, $GSI < 12$). The phonotactic responses of the gravid females consisted primarily of straight to slightly curved tracks to the sound source. Abrupt changes in trajectory were rarely observed (only three of the 45 positive responses). In most cases, females would contact the opaque tarp directly in front of the speaker and then proceed to swim under the tarp and make contact with the sound projector. A positive response was unambiguous and consisted of repeated contact with the speaker face and/or prolonged, active circling in front or underneath the sound projector. The majority of the gravid females in the test group [73%, 45 of 62 gravid females; size range: 9.0–12.8 cm standard length (SL), mean $SL = 11.0 \pm 0.7$ SD cm] responded to the 90 Hz playback tone and localized the sound source. In contrast, none of the gravid females in the control group ($n=59$, mean $GSI = 23.5 \pm 8.3$ SD, size range: 10.3–15.3 cm SL, mean $SL = 12.0 \pm 1.1$ SD cm) released with the sound off made physical contact or showed active interest in the silent projector. Instead, gravid females in the sound-off control group swam in many directions. Figure 8 shows the response pathways for both the test (sound-on) and control (sound-off) groups.

The initial bearing (the angle measured using the first two time points) of the control group did not show a directional preference [vector strength (VS)=0.157] and did not significantly differ from the null hypothesis of a random distribution of response angles ($p=0.24$, $n=59$). In contrast, the

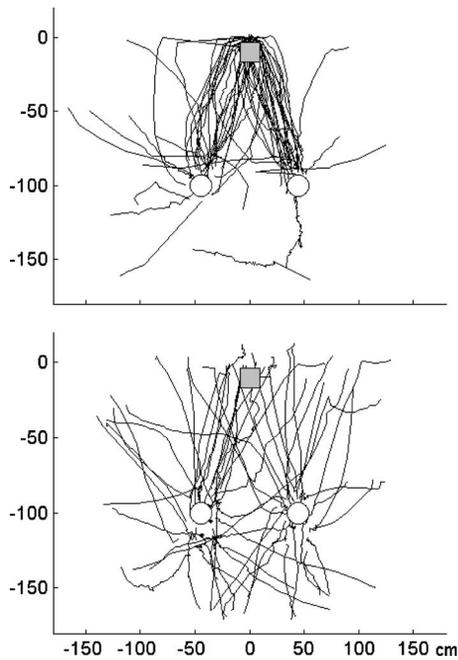


FIG. 8. Response pathways of both the test (sound on; upper panel) and control (sound off; lower panel) groups of naïve test animals. The axes are the distance from the center of the tank in cm. Note that the phonotactic responses of the gravid females in the test (sound on) group consisted primarily of straight to slightly curved tracks to the sound source.

initial bearing of the test group did show a directional preference toward the sound source (mean angle -9° , $VS = 0.462$), which differed significantly from random distribution of response angles ($p < 0.0001$, $n = 59$; note that three response pathways could not be analyzed from the video records because the fish swam under the release cylinder as it was lifted out of the tank and the pathway was visually obscured). Figure 9 shows scatter plots of the difference angles of the fish's bearing relative to the sound source (upper plot) and relative to the local particle motion vector (lower plot) as a function of distance to the sound source for the gravid females that exhibited positive phonotaxis. These plots indicate that the fish, on average, headed toward the source (0°) and in line with the particle motion vectors at every distance up to about 100 cm away from the source. In addition, positive phonotactic females showed a high degree of directional tendency ($VS = 0.7$ to 0.95) toward the sound source at distances from 80 to 0 cm away (Fig. 10). The high VS values reported at these distances indicate a robust directional preference with respect to both the source location and the acoustic particle motion vectors (p values < 0.0001 between 110 and 10 cm away from the sound source).

IV. DISCUSSION

Previous studies have shown that fish can orient to and locate sound sources. Many of these studies have used the playback of conspecific vocalizations to determine preferential attraction, directionality, and localization of sound sources (Tavolga, 1958; Winn, 1972; Popper *et al.*, 1973; Myrberg *et al.*, 1986; McKibben and Bass, 1998, 2001; Myrberg and Stadler, 2002; Rollo *et al.*, 2007; Rollo and Higgs, 2008). However, these previous playback studies lacked a

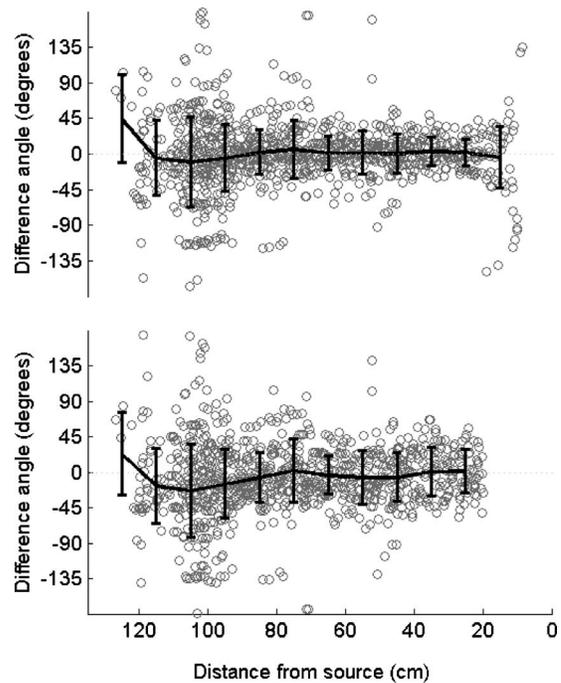


FIG. 9. Difference angles relative to the sound source as a function of distance to the source (upper panel) and the difference angles relative to vector field at 9.5 cm above tank floor as a function of distance to source (lower panel). The solid line and error bars in both panels are the mean and standard deviations.

full description of the sound field's structure. In this study the pathways that gravid female midshipman take as they approach a sound source were compared to the sound field, including the particle motion vectors, produced by that source.

A. Monopole sound field

The sound field produced by the J-9 sound projector in the behavioral arena was primarily monopole, in that the

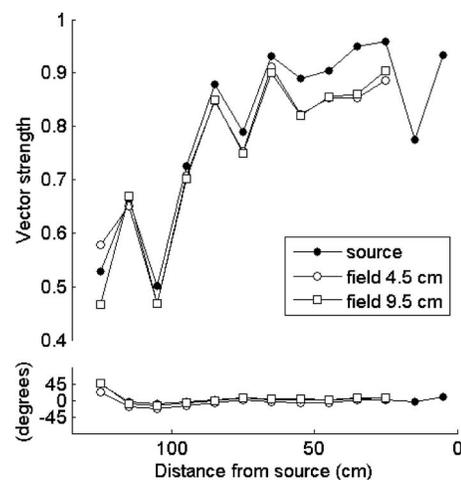


FIG. 10. Plots of vector strength (upper panel) and the mean difference angles (lower panel) as a function of distance to the sound source. Upper panel show vector strength measure relative to the sound source (\bullet) and relative to the local particle motion vector at 4.5 (\circ) and 9.5 cm (\square) above the tank floor. The p values were < 0.0001 between 110 and 10 cm away from the sound source, indicating the animals were directed toward the mean difference angle from those distances.

pressure decreased away from the source at about the same rate in all directions (Fig. 5) and the particle motion vectors pointed to (and away from) the source (Fig. 7). A monopole sound field was expected to be generated by this source but was not guaranteed considering the playback environment of the shallow test tank. Though minor, there was some local interaction with the bottom as the particle motion vectors at 9.5 cm above the bottom are more uniformly directed toward the source than the particle motion vectors at 4.5 cm above the bottom (Fig. 7). Also, closer to the tank wall the sound pressure begins to increase due to reflections from the wall, and the particle motion vectors do not necessarily point toward the source.

B. Phonotaxis and localization

As in previous midshipman playback studies (Winn, 1972; McKibben and Bass, 1998, 2001), only gravid females displayed a positive phonotactic response to a tone near the fundamental frequency of the male's advertisement call. It can be concluded from these experiments that gravid females can in fact localize sound sources and rely solely on acoustic cues to do so. The control (sound-off) group showed no directional tendency on initial release and none of the gravid females from the control group remained in the center of the tank (where the silent sound projector was located) if they happened to pass near it. However, upon release, the test (sound on) group showed significant directional tendency toward the source, and 45 of the 62 (73%) gravid females located the speaker and contacted the speaker face, and then remained under or circled the transducer as long as the sound remained on. McKibben and Bass (1998, 2001) demonstrated that gravid females swam to the source, but they were uncertain whether fish used only acoustic cues to locate the source. Here, only naïve fish were tested using one speaker that was visually occluded by an opaque screen. The only sensory cues available to the fish for localizing the sound source were acoustic.

C. Mechanisms and strategies of sound source localization

Investigators generally agree that particle motion provides information useful for determining the direction of a sound source, but it is presently a matter of debate (1) whether fish decide on the source location from a distance using local cues or use a strategy for approaching the source, (2) whether both particle motion detection and sound pressure detection are required for localization and resolution of the "180° ambiguity" (see below), and (3) whether the otolithic ears or the lateral line organs are the receptors responsible for localization behavior, particularly in the near field.

The majority of the gravid females (73%) exhibited a positive phonotactic response. Their pathways tended to be smooth, continuous, and consistently directed toward the source with comparable accuracy between the release site and final response at the source ($VS > 0.7$, Figs. 8 and 9). This is in contrast to sculpin that display a zig-zag pattern where they stop and seem to sample the environment and

then refine their behavior when approaching a nearby source using the lateral line system (Coombs and Conley, 1997). In the present study, midshipman did not adopt an arbitrary orientation and then swam in a direction that maintained a constant (but arbitrary) angle with respect to the axis of particle motion (*sensu* Kalmijn). Rather, they adopted (on average) a 0° orientation with respect to the particle motion axis, and swam toward the source. The finding that midshipman were significantly directed toward the sound source from the release sites indicates that a displacement of 45 dB re 1 nm (178 nm) is sufficient for these fish to determine direction.

It is possible that phonotaxis in the midshipman can be explained as the animal's orientation to the spatial change in intensity that exists in the tank (Fraenkel and Gunn, 1961; Richard, 1968), with the fish "climbing up" the intensity gradient. There is indeed an intensity gradient amounting to about 0.3 dB per cm (Fig. 5). Although such orientation strategies have not been demonstrated for fish experimentally, the performance in this task would presumably depend on the abilities of the fish to make comparisons of the sound levels at successive locations, and then to move in the direction of higher intensity. The discrimination limen for intensity has not been measured in the midshipman, but based on comparative data from other species, it is on the order of 2–9 dB (Fay, 1988). If the midshipman used their ears and not the lateral line for phonotaxis, they would presumably have to move in a zig-zag manner to sample the sound field, as anurans do (Rheinlaender *et al.*, 1979). The tracks taken by the animals in this study do not suggest this sort of behavior, and therefore, do not support a sampling and gradient climbing mechanism for source localization. If the lateral line system were used in this task, the pressure gradients appearing along the length of the animal could possibly be used for orientation if the intensity gradient acuity were adequate. However, this acuity has not been measured in any animal.

An important issue with respect to sound source localization by fishes concerns the "180° ambiguity" problem. Particle motion in a monopole sound field is ambiguous in that the axis of motion points both toward and 180° away from the source. Which direction should the fish swim? Theoretically, the phase relation between sound pressure and particle motion could be used to resolve the ambiguity (Schuijff, 1975), but it requires independent measurements of sound pressure and particle motion. Sensing sound pressure, presumably via the swim bladder, is a mode of hearing that is unlikely in midshipman and other toadfish, as there is no evidence so far for sound pressure reception in these animals. Nevertheless, further studies on midshipman phonotaxis using animals with deflated swim bladders are planned to investigate this possibility.

It is interesting to note that no evidence was found that the midshipman fish were confused by the 180° ambiguity of the particle motion. If sound source localization can take place independent of swim bladder involvement, then other strategies for resolving the 180° ambiguity must be available. It is possible that fish sample at two or more sites before deciding which direction to swim, or use estimates of the probabilities of source locations to influence their decisions as human listeners do when making sound source elevation

and front-back judgments in anechoic environments (Wightman and Kistler, 1993). In other words, it might be asking too much of the auditory system to be able to solve this problem in isolation.

Another important issue concerns the involvement of the lateral line system in near-field sound source localization. Up through the 1960s while the work of van Bergeijk (1964) was influential, it was thought that all near-field "hearing" by fishes was subserved by the lateral line system. It is now known that the otolithic organs of the ears are exquisitely sensitive to acoustic particle motion [with thresholds at 100 Hz on the order of 0.1 nm (Fay, 1984, 1988)], and that the lateral line system is a very close range detector (on the order of a few body lengths; Coombs and Conley, 1997). It remains a possibility that sound source localization behavior depends on lateral line stimulation, so phonotaxis experiments on midshipman for which the lateral line system of the midshipman has been inactivated are planned for future experiments.

V. SUMMARY AND CONCLUSIONS

In this study, the plainfin midshipman fish was used as a general model to explore how fishes localize an underwater sound source in the relatively simple geometry of a monopole sound field. It was found that gravid females were directed toward the sound source upon initial release and the majority (73%) subsequently followed straight to slightly curved pathways directed toward the source and in line with the local particle motion vectors. This study is the first to compare the path that fish take to a sound source with the local vector components of the sound field. It was not possible to conclude from these experiments if the fish "knew" where the sound source was located in the sense that humans (and other vertebrates) do, but it does appear that midshipman fish are able to use acoustic cues to determine direction to a sound source from distances of 80–100 cm away. There are a number of questions still to be answered, but the procedure presented here can be applied to more complex fields and represents an approach that may shed light on the cues and strategies that fish use to localize sound sources in their natural environment.

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