

RESEARCH ARTICLE

Local acoustic particle motion guides sound-source localization behavior in the plainfin midshipman fish, *Porichthys notatus*

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SUMMARY

Sound-source localization behavior was studied in the plainfin midshipman fish (*Porichthys notatus*) by making use of the naturally occurring phonotaxis response of gravid females to playback of the male's advertisement call. The observations took place outdoors in a circular concrete tank. A dipole sound projector was placed at the center of the tank and an 80–90 Hz tone (the approximate fundamental frequency to the male's advertisement call) was broadcast to gravid females that were released from alternative sites approximately 100 cm from the source. The phonotactic responses of females to the source were recorded, analyzed and compared with the sound field. One release site was approximately along the vibratory axis of the dipole source, and the other was approximately orthogonal to the vibratory axis. The sound field in the tank was fully characterized through measurements of the sound pressure field using hydrophones and acoustic particle motion using an accelerometer. These measurements confirmed that the sound field was a nearly ideal dipole. When released along the dipole vibratory axis, the responding female fish took essentially straight paths to the source. However, when released approximately 90 deg to the source's vibratory axis, the responding females took highly curved paths to the source that were approximately in line with the local particle motion axes. These results indicate that the acoustic cues used by fish during sound-source localization include the axes of particle motion of the local sound field.

Key words: phonotaxis, hearing, dipole source, fish, near field.

INTRODUCTION

Sound-source localization by fishes has been a topic of controversy since early negative results (e.g. von Frisch and Dijkgraaf, 1935) came to dominate expectations. It is now generally accepted that at least some species of fish can locate sound sources. The change in thinking has been slow and is the culmination of a long history of suggestive results (e.g. Popper et al., 1973), discrimination studies (e.g. Buwalda et al., 1983) and a few unequivocal demonstrations (e.g. Zeddies et al., 2010). One of the greatest challenges has been in understanding the sound fields within which fish solve the problems of localization, both in the production of the sound field and in its characterization. Reported here is the first attempt to create and measure the relatively complex sound field produced by a dipole sound source in a behavioral arena, and the demonstration of sound-source localization by fish in that arena.

Any sound radiator can be represented by a sum of a distribution of elemental sources, the simplest of which is a monopole. A dipole source can be created by two closely spaced monopoles that are out of phase or by a mono-axially translating rigid object. Although monopole radiation is omnidirectional (equal in all directions) (Fig. 1A), dipole radiation produces a bi-lobed, figure-eight pattern (Fig. 1B). The pulsating swim bladder of a fish likely acts as a monopole sound source and the body of a swimming fish may act as a dipole radiator (e.g. Kalmijn, 1988).

An acoustic disturbance can be described at a point in space by the pressure (scalar) and particle motion (vector). In the near field of a monopole sound source, pressure and particle motion spatially decay at different rates, independent of angle. The axes of particle motion in the omnidirectional, monopole field all point towards or away from the source (Fig. 1C). For a dipole, the decay rate of the pressure and particle motion and the relative direction of the particle motion axes are a function of location within the field. Along the dipole axis, i.e. in the direction of the axis connecting the two appropriately placed elemental monopoles (or along the translation axis), the pressure and particle motion decay in a manner similar to that of the monopole (though at different rates) and the particle motion axes point towards or away from the source (Fig. 1D). However, in the direction orthogonal to the dipole axis, the sound pressure is nulled (Fig. 1B) and the particle motion axes are parallel to the dipole axis (Fig. 1D). At intermediate bearings, the particle motion direction gradually changes from parallel to the dipole axis near the pressure null to pointing towards or away from the source along the dipole axis. In other words, the particle motion axes surrounding a dipole do not necessarily point towards or away from the source as they do for monopoles.

It is now generally accepted that for fishes to localize sound sources they must first detect and determine the axes of particle motion using their otolithic ears [vector detection (Schuijf, 1975)].

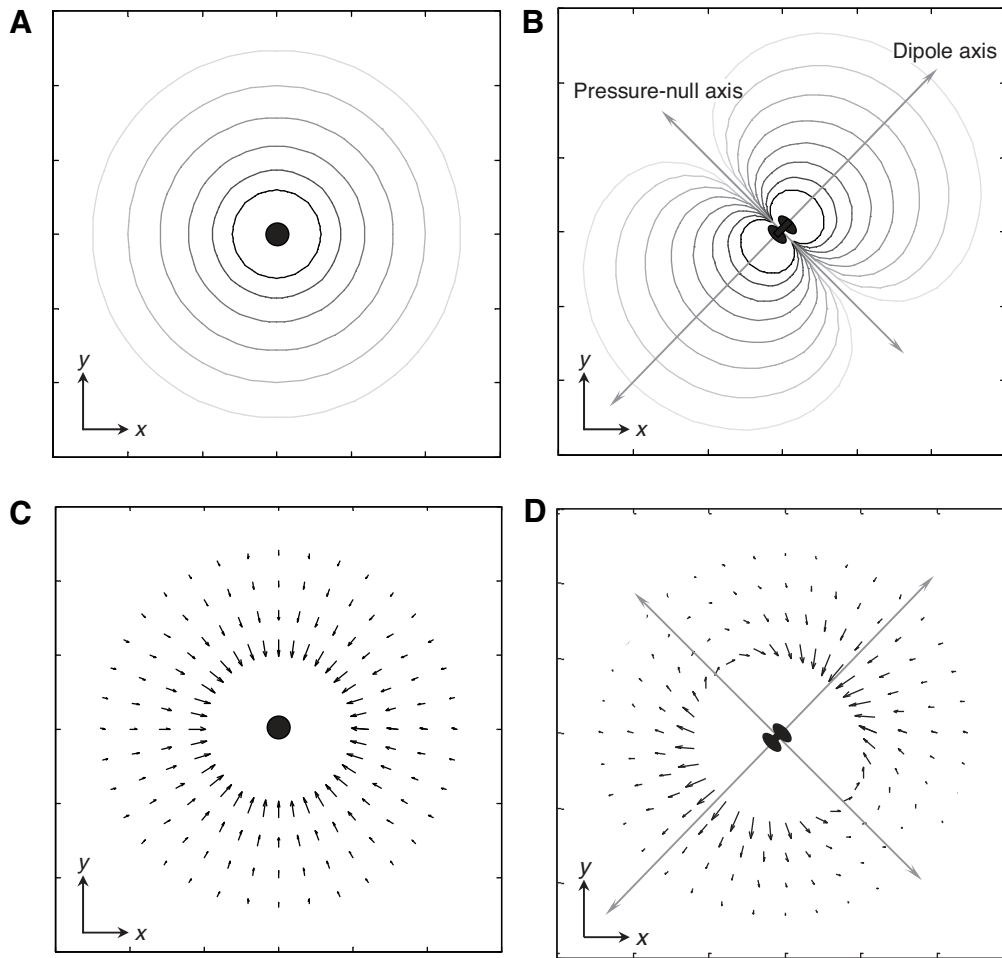


Fig. 1. Two-dimensional spatial projections of acoustic pressure and acoustic particle motion fields for an ideal monopole projector and an ideal dipole projector. (A) Contour plot of the pressure field surrounding an ideal monopole sound projector. Pressure magnitude is denoted by darkness of the contour band, with higher pressure being darker. The pressure field is radially symmetric, or omnidirectional, with respect to the projector. (B) Contour plot of the pressure field surrounding a dipole projector. The pressure field is bi-lobed with areas of high pressure along the dipole axis, and a pressure null orthogonal to the dipole axis. (C) Particle motion vectors surrounding a monopole source. The particle motion field is radially symmetric field, with all of the particle motion vectors pointing towards (or away from) the source. (D) Particle motion vectors surrounding a dipole source. Particle motion vectors along the dipole axis point towards (or away from) the source, whereas particle motion vectors along the pressure-null axis are parallel to the dipole axis. In addition, moving from the pressure-null to the dipole axis, the particle motion direction gradually changes from parallel to the dipole axis to pointing towards or away from the source.

A dipole source has seldom been used in sound-source localization experiments with fish, but dipoles may be a common type of source in nature (Kalmijn, 1988) and offer the opportunity to observe behavior when fish are in locations where particle motion axes do not point towards or away from the sound source. In the present experiment, we continue to use the plainfin midshipman (*Porichthys notatus*) as a model to explore how fishes localize underwater sound sources (Zeddies et al., 2010) and here we investigate how fish localize a more complex dipole sound source. We describe the phonotactic pathways of reproductive females that localize a dipole sound source when: (1) the fish were released along the dipole axis where the sound pressure is high and particle motion vectors point to or from the source, and (2) when the fish were released at a point along a line orthogonal to the dipole axis where the sound pressure is low and the particle motion vectors do not point to or from the source. Because the local sound field differs at the release sites, it was hypothesized that the pathways the fish would take to the source from these release sites would also differ if the local axis of particle

motion was the crucial sensory cue that fish use for locating sound sources.

MATERIALS AND METHODS

Experimental animals

Ninety-seven adult female plainfin midshipman fish, *Porichthys notatus* Girard 1854, were collected during the summer reproductive season (June and July 2009). Fish were collected during low tide from the nests of type I males within the intertidal zone of Tomales Bay near Marshall, CA, USA, the same geographical location used in our previous study (Zeddies et al., 2010). Reproductive females were readily distinguished from nesting (type I) and sneaker (type II) males based on the size of the animal and the shape of the urogenital papilla. Reproductive (gravid) females were also visually distinguished from type I males based on the size and appearance of the abdomen, which were typically distended because of the presence of eggs in gravid females and noticeably flaccid in spent females (Brantley and Bass 1994; Bass, 1996). All animals were

collected in the field during the early morning low tides and housed for a few hours in coolers with aerated seawater until they could be brought to the Bodega Marine Laboratory (BML) in Bodega Bay, CA, USA. At BML, the animals were maintained in large communal aquaria at natural ambient temperatures (12–14°C) until tested later that night. All experimental procedures were approved by the University of California Davis Institutional Animal Care and Use Committee.

Experimental tank and setup

All tests were conducted outdoors [as in Zeddies et al. (Zeddies et al., 2010)] in a cylindrical concrete tank (4 m diameter, 0.75 m depth) at BML. A ‘dipole’ sound source was constructed by connecting two underwater loudspeakers (Lubell AQ339, Clark Synthesis, Littleton, CO, USA) back to back and driving them in opposite phase. The combined sound source was suspended from a beam so that it was at the center of the tank and the center of the projector face was positioned 10.5 cm above the tank floor. The axis connecting the two projector components is shown in Fig. 2. To remove possible visual cues that might affect sound localization behavior, we used a 2.44 m opaque plastic tarp as a screen that was placed immediately in front of, but not touching, the sound projector.

Four release sites were used (labeled A, A', B and B' in Fig. 2). Release sites A and A' were approximately on the dipole motion axis 79 and 98 cm away from the sound source, respectively, whereas B and B' were approximately perpendicular to this axis at 79 and 98 cm away from the sound source, respectively. Both near and far release positions were used in these experiments to maximize the probability of the fish responding to the sound playback stimuli.

The playback signal consisted of a continuous tone at either 80 or 90 Hz. The playback signal was similar to the fundamental frequency of the male advertisement call [80–100 Hz (Bass et al., 1999)], and we used either 80 or 90 Hz depending on the water temperature in the behavioral arena on the night of the experiment.

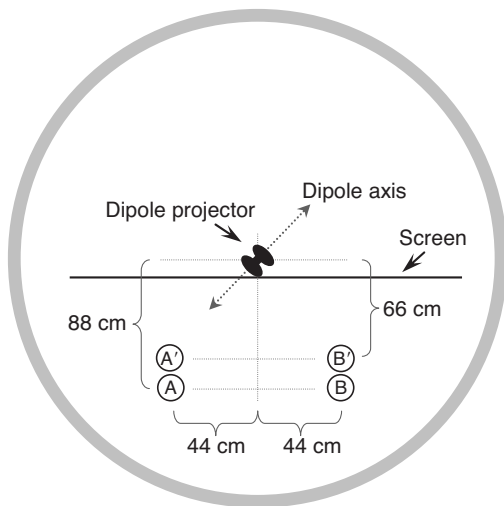


Fig. 2. Schematic of experimental setup (tank diameter=4 m; water depth=50 cm). The dipole source consisted of two Lubell AQ339 projectors rigidly attached along their central axis and driven in opposite phase. The center of the combined projector was placed at the center of the tank. A, A', B and B' indicate the animal release sites; note that A and A' are along the axis of the dipole source whereas B and B' are approximately perpendicular to the dipole axis.

McKibben and Bass showed previously that females exhibit a temperature-dependent frequency preference for approaching a sound source that mimics the fundamental frequency of the male's advertisement call (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, USA) that drove the sound projector. The sound level at release site A (Fig. 2) was calibrated nightly using a custom-written LabVIEW program with a National Instruments DAQPad-6052E (Austin, TX, USA) and a mini-hydrophone (8103, Brüel and Kjær, Norcross, GA, USA) placed 4.5 cm above the tank floor. The tone level at the calibration site was 120 dB (re. 1 µPa) 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 using a digital video recorder and a CV110 Precision black-and-white camera (0.21x minimum light level; Keyence, Mechelen, Belgium) 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, USA) and iMovie 7.0 software (Apple, Cupertino, CA, USA). Windows Movie Maker 5.0 (Microsoft, Redmond, WA, USA) and SigmaScan Pro 5.0 (Systat Software, Inc., Chicago, IL, USA) 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.

Experimental protocol

The sound playback experiments were conducted at night between 21:00 and 03:00 h during the midshipman summer breeding season in June and July 2009. Three red floodlights positioned around the tank perimeter allowed us to observe and videotape the behavioral responses of female midshipman fish to the acoustic playback stimuli. The water temperature in the test tank (range=10–12°C) 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 and 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 when an individual fish was placed in a 30 cm diameter plastic mesh cylinder at release site A, A', B or B' while the sound playback stimulus was playing (see Fig. 2). 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 (McKibben and Bass, 1998) and was used because it increased the likelihood of a positive response in previous experiments. 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 ultimately approached the sound source and then directly touched the speaker face or circled in front of or under the dipole sound projector.

Acoustic and vibration measurements

Acoustic pressure measurements were made on a Cartesian grid throughout the tank at 9 cm increments in the front half of the tank (the half with the release sites) and at 22.5 cm increments in the

back half of the tank. The pressure measurements were obtained at 9 cm above the tank floor using miniature (50 mm length, 9.5 mm diameter) Brüel & Kjaer type 8103 hydrophones. These hydrophones have a useful frequency range of 1 Hz to 170 kHz and a sensitivity of approximately -212 dB re. $1 \text{ V } \mu\text{Pa}^{-1}$. The output signals from the hydrophones were amplified by charge amplifiers (Brüel & Kjaer model 2692) and fed into a DAQPad-6052E. Custom LabVIEW programs were written to control and record from the DAQPad. The pressure calibration of each hydrophone was tested with a Brüel & Kjaer 4229 pistonphone calibrator. All hydrophones measured within 0.25 dB of the expected voltage.

To measure acoustic particle motion, a probe consisting of a pair of hydrophones bounding a triaxial accelerometer was created (Fig. 3). The accelerometer (PCB model W356A12, sensitivity = 100 mV g^{-1} ; PCB Piezotronics, Inc., Depew, NY, USA) was made neutrally buoyant in water by embedding it in a syntactic foam enclosure. All three sensors were held in place within a flooded, PVC-tube frame using a compliant natural rubber suspension, and arranged such that their centers of sensitivity were co-linear. The validity of the responses of the sensors in the probe frame was confirmed through comparison with responses of individual sensors obtained without the probe frame.

At each probe location within the water tank, data from all three sensors were collected simultaneously using a Measurement Computing PCI-DAS6052 data acquisition board (Measurement Computing Corporation, Norton, MA, USA) controlled by a custom graphical interface operating in MATLAB (MathWorks, Natick, MA, USA). The data collection system generated the dipole drive signal and created the trigger for sensor data acquisition, allowing consecutive signals to be coherently averaged. Ten to 20 signal ensembles were averaged in order to improve measurement signal-to-noise ratios. This was crucial given the high rate of spatial decay in the extreme near field of the source. Each probe sensor data record was 1.0 s in duration, and was sampled at 55,096 Hz.

A radial arm with its fixed rotation point vertically above the dipole source (center of the tank) was created so that the probe could be positioned along radii directed away from the source. A high-frequency acoustic locator system was installed to measure the location of the probe frame during the field scans. The in-water

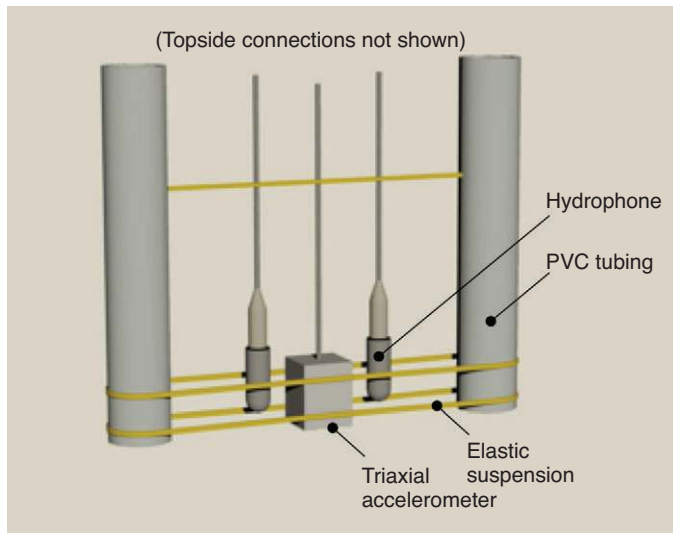


Fig. 3. Probe arrangement to measure particle motion. A triaxially accelerometer (PCB model W356A12) was suspended in a flooded PVC tubing frame using natural rubber elastic. Two hydrophones (Brüel & Kjaer 8103) were also mounted so that they were collinear to the accelerometer.

system consisted of three $\frac{3}{4}$ -inch diameter piezoelectric spheres, each of which could transmit or receive. One was fastened to the probe arm and functioned as a transmitter. The other two spheres functioned as receivers and were fastened to flooded PVC tubes along the fixed beam from which the dipole was suspended. The transmitter sphere was driven with a short broad-band pulse centered at 35 kHz, and the radiated signals were received on the two stationary spheres. The pinger drive signal and trigger were generated by the data acquisition system using a second board with an output clock rate of 330 kHz. The two pinger receiver signals were recorded on this board at a sampling rate of 165 kHz. The times of flight between the source and receiver pingers were calculated using conventional replica-correlation processing. Using the sound speed in water and independent measurements of the transducer geometry, time of flight calculations were then converted to estimates of distance between the probe and the projector, and the angle the probe arm made relative to the projector suspension beam. Acoustic acceleration measurements were obtained, for the front half of the tank that included the release sites, at approximately 10 deg angular increments and 10 cm radial increments. The precise locations of the measurements sites were then determined by the high-frequency locator system. The precision of the calculations was approximately 0.9 mm, but the accuracy of the calculations was approximately 5 mm, owing to uncertainties in the absolute locations of the pinger transducers.

Acoustic modeling

An analytical model was developed to compute estimates of pressure and particle velocity sound fields for a dipole sound source placed at the center of the tank. The dipole source was modeled as an offset pair of identical, finite-sized, oppositely phased radiators. Based on separate observations (Alteirac, 2001), the volume velocity distribution of each radiator was modeled to have a maximum value at the geometric center, and tapering to zero at the edges with a Hanning window. The effects of the finite water column and its boundaries were accounted for by representing the dipole volume velocity distribution in terms of duct modes, assuming a rigid bottom and a pressure-release water-air interface. The modes are of the form $\sin[(2n-1)\pi z/2H]$, where n is an integer greater than or equal to 1, z is the vertical coordinate and H is the water depth [see p. 314 in Pierce (Pierce, 1989)]. The model captures the extreme near field of a dipole projector in an asymmetric waveguide at frequencies where the water column is acoustically small (i.e. the wavelength of sound is much longer than water depth: $k_0 H \ll 1$, where k_0 is the acoustic wavenumber). It was found that the spatial decay rates were sufficiently high that effects of the tank side walls could be neglected in the field predictions for the range of locations relevant to the behavioral testing.

For each location of interest in the tank, field pressures were calculated at five total locations: the location itself, two points laterally offset by ± 1 mm, and two points radially offset by ± 1 mm. The pressures at these offset points were used to calculate the tangential and radial particle velocities using Euler's equation, which relates pressure and velocity in a fluid in the absence of viscous and gravitational effects:

$$\mathbf{v}_r = \frac{1}{i\omega\rho} \frac{[p(r-\Delta) - p(r+\Delta)]}{2\Delta}, \quad (1)$$

and

$$\mathbf{v}_t = \frac{1}{i\omega\rho} \frac{[p(r\theta-\Delta) - p(r\theta+\Delta)]}{2\Delta}, \quad (2)$$

where v_r and v_t are the radial and tangential particle velocities, respectively, i is the square root of -1 , ω is the angular frequency, ρ is the density of water, p is the pressure, r is the radial distance of the location, θ is the angle to the location relative to the dipole axis and Δ is the radial (for v_r) or tangential (for v_t) position offset from the location at which the velocities are sought.

Analysis of movement

The difference angles of the bearing of the fish relative to the source and relative to the particle motion axes of the local sound field were determined as the fish left the release sites (Fig. 4). This was accomplished by determining the position of the fish in two consecutive video frames as the fish moved away from the release sites. 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 calculated from the model at the fish's position.

The difference angles relative to the source *versus* relative to the local sound field by the fish released at sites A and B were compared using a one-way repeated-measures ANOVA followed by the Newman-Keuls *post hoc* comparison test for planned comparisons. For all tests, α was set at 0.05. Statistical analyses were performed using the software program STATISTICA for Windows (StatSoft, Inc., Tulsa, OK, USA).

RESULTS

Acoustic pressure and particle motion measurements

In order to evaluate the phonotactic behavior of female midshipman, we first characterized the sound field of the acoustic playback stimuli [80–90 Hz, the approximate fundamental frequency of the male advertisement call, adjusted to match the appropriate temperature (see McKibben and Bass, 1998)] that was produced using a dipole sound projector. Quantitative descriptions (maps) of the acoustic

pressure and particle motion fields in the behavioral arena were recorded using the hydrophone/accelerometer probe.

The sound pressure field encountered by fish during behavioral experiments was mapped at 7 cm above the bottom of the tank at a resolution of 9 cm in the front half of the tank (where the release sites were located) and 22.5 cm in the back half of the tank. Fig. 5 shows a contour plot of the sound pressure field at 80 Hz. The sound pressure field is that expected of a dipole source. It shows a bi-lobed field with high sound pressure in line with the dipole axis, and low pressure (null) perpendicular to the dipole axis. Release sites A and A' were approximately along the dipole axis, whereas release sites B and B' were approximately along the pressure-null axis.

Particle motion in the front half of the tank was measured using a probe consisting of a triaxial accelerometer and two hydrophones (Fig. 3). Proper description of particle motion is a vector in three-dimensional space. However, for ease of presentation, two-dimensional vectors in the x - y plane (parallel to the bottom of the tank) are shown (Fig. 6). Fig. 6 also includes modeled predictions of particle velocity for an ideal dipole source in this tank. The modeled results agree with the measured particle velocities and support the conclusion that the source was acting as a dipole in the tank.

It can be seen in Fig. 6 that at release sites A and A', the particle motion vectors point towards the projector (or away: arrowheads were added arbitrarily to only one end of the vector). Whereas, at release sites B and B', the particle motion vectors are approximately parallel to the dipole axis, and perpendicular to the line towards the projector from the release site. The magnitude of displacement was 17 and 20 dB re. 1 nm at release sites A and A', respectively, and 17 and 18 dB re. 1 nm at release sites B and B', respectively.

Phonotactic response pathways

A positive phonotactic response was unambiguous and resulted in repeated contact of the fish with the speaker face and/or prolonged,

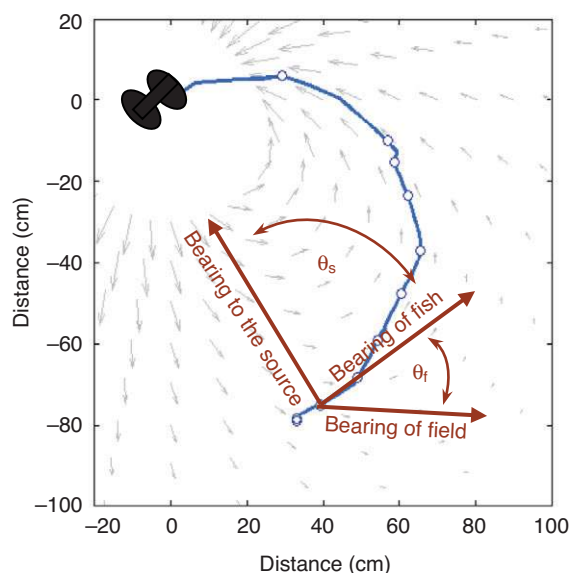


Fig. 4. Difference angle calculation. The difference, or error, angle that the fish took when leaving the release sites was determined by calculating the initial bearing that fish swam relative to the local particle motion bearing and relative to the bearing towards the sound source.

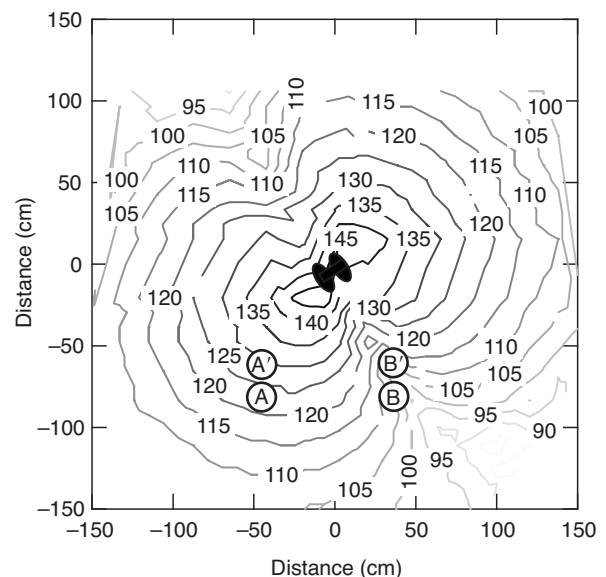


Fig. 5. Sound pressure field in the behavioral arena at 80 Hz. The dipole projector is shown in the center of the tank, and the location of the release sites A, A', B and B' are shown. The sound pressure levels are in dB re. 1 μ Pa.

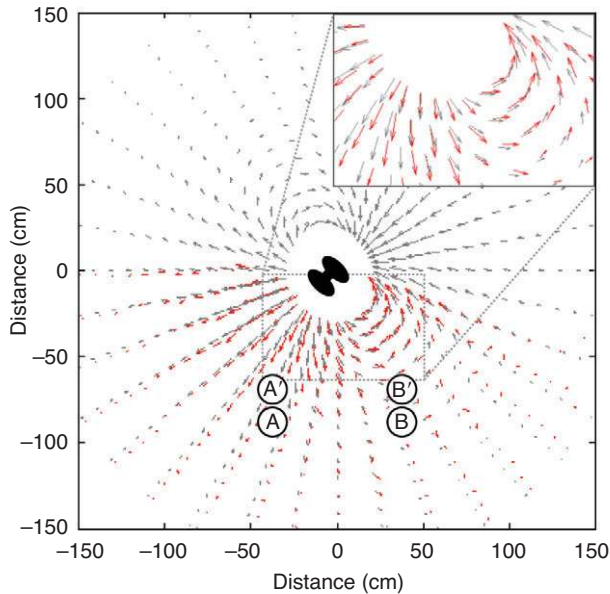


Fig. 6. Acoustic particle motion fields in the behavioral arena. The dipole projector is shown at the center of the tank and the release sites A, A', B and B' are shown. Red arrows are the particle velocity vectors measured using an accelerometer. Gray arrows are the particle velocity vectors generated by the model. The inset shows close correspondence between the measured and modeled vectors. Measurement locations were determined with a high-frequency acoustic locator system (see Materials and methods); model locations were at regular intervals along radials. The heads of the arrows indicate one phase only, namely compression of the left/lower projector and rarefaction of the right/upper projector.

active circling in front of or underneath the sound projector. Positive phonotactic responses were only observed in gravid females (full of eggs) whereas recently spawned or 'spent' females (void of eggs; $N=13$) never exhibited phonotaxis or resulted in contact with the sound projector. Of the 84 gravid females collected, 52% ($N=44$) exhibited positive phonotactic responses. The pathways of these gravid females ($N=44$) released from the four release sites that responded to the dipole sound source are shown in Fig. 7. At release sites A and A', the phonotactic responses of gravid females ($N=20$) consisted primarily of straight to slightly curved tracks to the sound source. These pathways are essentially the same as those reported earlier (Zeddies et al., 2010) for a monopole source. The pathways of gravid females ($N=24$) released at sites B and B' are quite different from those at A and A'. They consist of approximately equal numbers that exit the release sites to the left ($N=10$) and to the right ($N=7$), and the pathways to the source are generally curved and appear to follow the local particle motion vectors (Fig. 7). The majority of fish (16/24 or 67%) followed the particle motion vectors from the release site (Fig. 8A) whereas others moved a short distance (~5–10 cm) and then began to follow the particle motion vectors ($N=3$; Fig. 8B); some ($N=5$; Fig. 8C) approached the sound source directly and did not follow the particle motion vectors.

The angles that the fish leave the release sites were compared. The initial bearing of each fish (Figs 7 and 8, dark arrows) was used to define the orientation error with respect to the bearing towards the source, and with respect to the bearing of the particle motion vectors calculated by the model at the location of the fish. At release sites A and A', the direction towards the source and the particle motion vectors are similar, so it was hypothesized that animals

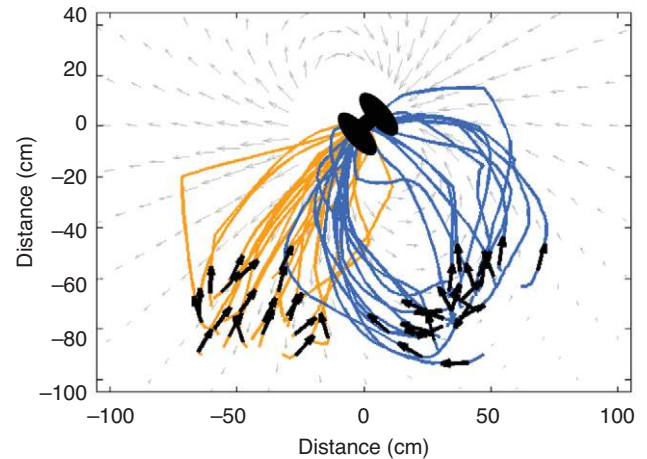


Fig. 7. Response pathways of the naïve fish as they approach the sound source. Orange traces are fish released from sites A and A'; blue traces are fish released from sites B and B'. Gray arrows indicate the particle velocity vectors generated by the model. Black arrows are the initial directions of the fish as they leave the release sites.

released at sites A and A' would have a similar and small error with respect to the source and the particle motion vectors. At release sites B and B', however, the direction towards the source and the particle motion vectors were nearly perpendicular. Therefore, it was hypothesized that the orientation error relative to either the source or the particle motion would be similar to the error at A and A', and the other would be significantly different. We found that there were no differences in orientation angle errors from fish released at sites A or A' with respect to the source (mean absolute orientation error=17 deg) versus the local particle motion vectors (mean absolute orientation error=19 deg). There were significant differences in the orientation angle errors between fish released at sites B or B' with respect to the source (mean absolute orientation error=51 deg) versus the local particle motion vectors (mean absolute orientation error=28 deg) (one-way repeated-measures ANOVA, Newman-Keuls *post hoc* comparison test, $F=10.16$, $d.f.=3$, $P<0.001$). In addition, there were no differences in orientation angle errors of the fish with respect to the local particle motion vectors released at sites A or A' versus B or B' ($P=0.20$), but there were significant differences in orientation angle errors of fish with respect to the source released at A or A' versus B or B' ($P<0.001$). Clearly, the pathways from release sites B and B' are more in alignment with the local particle motion vector angles than they are with the direct path angle to the source.

The angles the fish took relative to the direction of the local sound field (particle motion vector) and relative to the sound source were calculated at each point as the fish approached the sound source (Fig. 9). The difference angle of the bearing of the fish relative to the sound field or the source from release sites A and A' (Fig. 9A,C) is, on average, approximately zero. The difference angle fish took from release sites B and B' relative to the local sound field vectors is also approximately zero (Fig. 9B). However, the difference angle fish took relative to the source from release sites B and B' (Fig. 9D) deviated from zero, and this deviation was most pronounced near the release site. These data are consistent with the hypothesis that local particle motion is an important cue that guides sound-source localization behaviour in the midshipman fish.

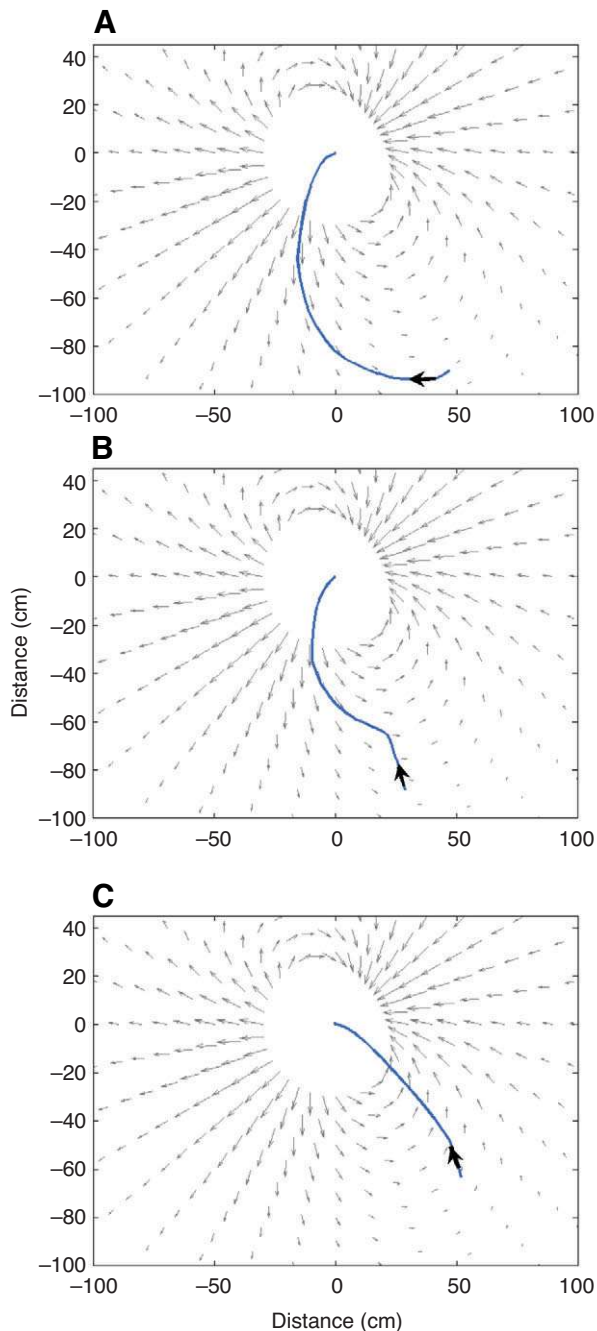


Fig. 8. Examples of pathways from release site B. (A) Example of a fish leaving site B that appears to follow the particle motion vectors from the release site. (B) Example of a fish that leaves site B and then abruptly changes course to align with the particle velocity vector field. (C) Example of a fish that goes approximately directly towards the source and does not follow the particle velocity vectors.

DISCUSSION

Previous 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; McKibben and Bass, 2001; Myrberg and Stadler, 2002; Rollo et al., 2007; Rollo and Higgs, 2008; Zeddies et al., 2010). No prior studies, however, have observed phonotaxis in the complex sound field of a known

dipole source. In this study we created and measured a dipole sound field and compared the pathways that gravid female midshipman fish take as they approach the sound source. The errors as the fish moved away from the release sites relative to the local particle motion vectors and relative to the direction toward the source were calculated. It was found that midshipman fish tend to follow the axes of the local acoustic particle motion vectors produced by the dipole source, regardless of whether these vector angles pointed towards the source.

Dipole sound field

The sound field produced by the sound projector in our tank showed that the distribution of sound pressure and acoustic particle motion in the test arena had the complex spatial distributions expected for a dipole source. The sound pressure distribution was substantially axi-symmetric with a relative pressure null at locations perpendicular to the vibrational axis of the source (Fig. 5), and the particle motion distribution had a relatively large amplitude with motional vectors that were nearly perpendicular to the line to the source at this same location (Fig. 6). These are the hallmarks of a dipole field.

Phonotaxis and localization in a dipole field

We conclude from these experiments that gravid females can use acoustic cues to localize dipole sound sources. We assume that all midshipman fish (and probably most other species) can locate sound sources as demonstrated here, but only gravid female midshipman are motivated to approach the sound source. When released at a location along the dipole axis (A and A'), the fish swam essentially directly towards the source along pathways parallel to the axes of particle motion at this location in the field (Fig. 7). This result replicates those reported previously (Zeddies et al., 2010) using a monopole source, where the fish swam in the direction of the local particle motion axes and along the axis pointing at the source, regardless of the point in the field at which the fish were released. However, when released at a location perpendicular to the dipole axis (B and B'), the fish swam (primarily) along curved pathways to the source. These curved pathways are consistent with the bearing of the local particle motion axes, but are not consistent with the bearing towards the source (Fig. 7). Therefore, in all cases (and we assume for all sources), midshipman fish tend to follow the axes of local particle motion.

In the wild it is unlikely that midshipman fish ever encounter a dipole version of the male's advertisement call. Although there may be some evidence for quadrupole projection very near the animal (Fine et al., 2001), the size of the swim bladder is small relative to the wavelength of sound at the call frequencies, meaning that the swim bladder would primarily radiate as a monopole and not as a dipole. Thus, a curved pathway to the source is unlikely. Nevertheless, the midshipman fish in this experiment were as likely to follow a curved path as a straight path to the source. In this sense, the local particle motion cues appear to determine the fish's behavior at each point along the path taken.

Mechanisms and strategies for sound-source localization

Gravid midshipman fish exhibiting a positive phonotactic response had approach pathways that tended to be smooth, continuous and at a fixed orientation angle with respect to the axes of local particle motion (Fig. 7). In this sense, midshipman could be behaving according to a strategy analogous to the 'light compass reaction' (Fraenkel and Gunn, 1961), in which the local particle motion vectors play the role of the sun, to which the animal maintains a constant orientation angle in order to reach the goal. This is

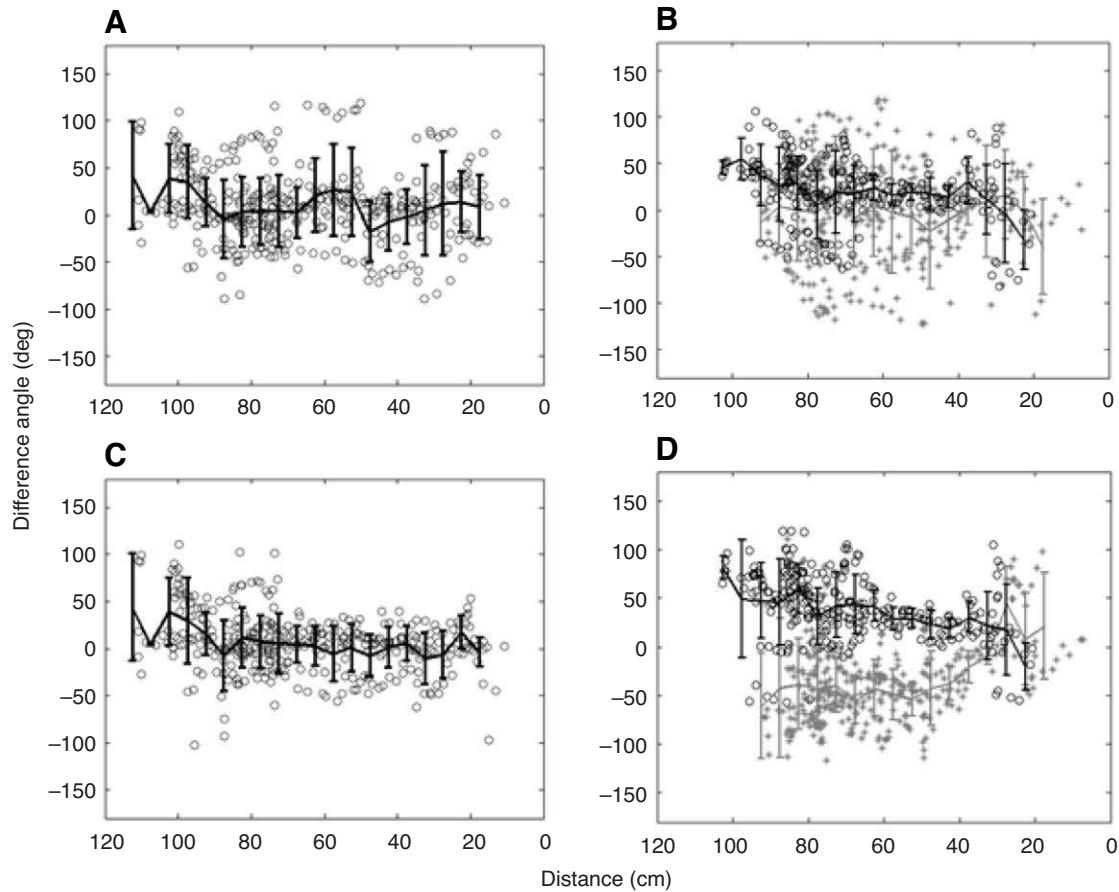


Fig. 9. Angles of fish movement relative to the sound field or the source as the fish approach the source. (A) Angles the fish take relative to the direction of the local sound field direction when released from site A. (B) Angles the fish take relative to the local sound field direction when released from site B. Because of the inherent 180 deg ambiguity in the direction of the local sound field vector, a positive response from release site B could be obtained by following the local sound field direction either to the right or to the left. Because of this ambiguity, the fish were grouped by those leaving towards the left (open circles) or the right (crosses). (C) Angles the fish take relative to the source when released from site A. (D) Angles the fish take relative to the source when released from site B. Again, because of the 180 deg ambiguity, the fish were grouped by those leaving towards the left (open circles) or the right (crosses). The angle the fish take relative to the sound field or the source from release site A (A,C) is, on average, approximately zero, as is the angle the fish take from release site B relative to the sound field (B). The angle the fish take relative to the source from release site B (D) deviates from zero, and the deviation is most pronounced near the release site

essentially the hypothesis of Kalmijn (Kalmijn, 1997), who recognized that in sound-source localization, a fish adopting any constant orientation angle with respect to the particle motion vectors would be using a strategy that would lead to the nearby acoustic source. However, the present results indicate that the adopted orientation angle is approximately 0 deg, and not an arbitrary angle. Thus, phonotaxis in the midshipman is more like a tropo-taxis response (Fraenkel and Gunn, 1961) than a light compass-like reaction. The present results demonstrate that for a dipole source, the animal does not ‘know’ where the source is, but can behave in a manner (by adopting a 0 deg orientation angle relative to the local particle motion vector) that results in reaching the source. This is not evident from experiments using a monopole source because adopting a 0 deg orientation angle predicts the same pathways as ‘knowing’ where the source is.

The behavior of midshipman is in contrast to the lateral-line-mediated orientation of sculpin that display a zig-zag pattern (klinotaxis) where they stop and seem to sample the environment and then refine their behavior when approaching a nearby source (Coombs and Conley, 1997).

The roles of the inner ear and lateral line system in near-field sound-source localization remain unresolved. Harris and van Bergeijk (Harris and van Bergeijk, 1962) first suggested the lateral line’s involvement in near-field sound source localization, and, van Bergeijk later posited that the lateral line and the inner ear should be thought of as parts of an ‘acoustico-lateralis’ system that could compute sound-source location within the near field (van Bergeijk, 1964). van Bergeijk assumed that the inner ears of fishes could only detect sound pressure (*via* the swim bladder ear linkage) and discounted their utility in detecting acoustic particle motion. Instead, van Bergeijk (van Bergeijk, 1964) argued that all near-field ‘hearing’ by fishes was subserved by the lateral line system. It is now known that the inner-ear, otolithic endorgans of fishes are exquisitely sensitive to acoustic particle motion, with thresholds on the order of 0.1 nm (Fay, 1984; Fay, 1988), and can be used to detect particle motion stimuli in the near field (and the far field, given sufficient particle motion amplitude). Recent, studies have shown that (vibrating) dipole sources robustly stimulate the inner ear in both pressure-sensitive fish (Coombs, 1994; Dailey and Braun, 2009; Nauroth and Mogdans, 2009; Dailey and Braun, 2011) and non-

pressure-sensitive fish (Coombs, 1994; Casper and Mann, 2007; Braun and Coombs, 2010). Although no major theories of sound-source localization require interactions between the inner ear and the lateral line, localization is still thought to result from multimodal integration within the octavolateralis systems (Braun et al., 2002). Both the inner ear and the lateral line system could play a role in midshipman sound localization behavior when the animal is close to the source, but their relative contributions are yet to be determined. Future studies of midshipman phonotaxis using animals in which pressure sensitivity or the lateral line system has been eliminated could be instrumental in determining the contributions of the inner ear and the lateral line to near-field sound-source localization.

Conclusions

In this study we used the plainfin midshipman fish as a general model to explore how fishes localize an underwater sound source in the relatively complex geometry of a dipole sound field. We found that gravid females were directed towards the sound source upon initial release, and that they followed nearly straight pathways to the source when released along the dipole axis (release sites A and A') and highly curved pathways to the source when released perpendicular to the dipole axis (release sites B and B'). From all release sites, the fish swam in line with the local particle motion vectors. It is now clear that for dipole fields, midshipman fish do not appear to 'know' where the source is, but can use acoustic cues of local particle motion vectors to determine the direction to swim in order to reach the sound source.

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