Chapter 25
From Cave Fish to Pile Driving: A Tail of Fish Bioacoustics

Arthur N. Popper

Arthur N. Popper

> Fishes have neither organs of hearing, nor yet the exterior orifice. And yet, it is quite certain that they do hear; for it is a well-known fact that in some fish-ponds they are in the habit of being assembled to be fed by the clapping of hands.
>
> —Pliny the Elder (about 50 CE), 1890 edition, p. 547[1]

[1] Or, if one wants the original Latin: "*Pisces quidem auditus nec membra habent nec foramina*, *audire tamen eos palam est*, *utpote cum plausu congregari feros ad cibum adsuetudine in quibusdam vivariis spectetur et in piscinis Caesaris genera piscium ad nomen venire quosdamve singulos. itaque produntur etiam clarissime audire mugil*, *lupus*, *salpa*, *chromis et ideo in vado vivere*." (From http://penelope.uchicago.edu/Thayer/L/Roman/Texts/Pliny_the_Elder/10*.html – see part LXXXIX).

A.N. Popper (✉)
Department of Biology and Center for Comparative and Evolutionary Biology of Hearing, University of Maryland, College Park, MD 20742, USA
e-mail: apopper@umd.edu

A.N. Popper and R.R. Fay (eds.), *Perspectives on Auditory Research*, Springer Handbook of Auditory Research 50, DOI 10.1007/978-1-4614-9102-6_25,

25.1 A Bit of History

Interest in fish sounds and hearing goes back about 2000 years to its first mention by the ancient Roman Pliny (the Elder). However, the first experimental studies of fish hearing did not come until the early 20th century with work by such prominent investigators as G. H. Parker (1902), Karl von Frisch (1923), and von Frisch's student Sven Dijkgraaf (e.g., 1932).

The "modern era" in studying fish hearing can be clearly linked to the classic and pioneering study by Tavolga and Wodinsky (1963). In this study, Bill Tavolga (Fig. 25.1) and Jerry Wodinsky, working at the Lerner Marine Laboratory in Bimini, the Bahamas, used psychophysics to measure the hearing sensitivity in nine species of marine fishes. Tavolga and Wodinsky trained fish to swim over a barrier in a "shuttlebox" to avoid a mild electric shock (something that might not get through an animal study regulatory committee today!) (Fig. 25.2a). The results of the study were the first comparative psychophysical hearing data for fishes, and the first to give accurate and repeatable thresholds and fish hearing ranges (Fig. 25.2b).

Although anyone can read this classic paper (available at http://digitallibrary.amnh.org/dspace/handle/2246/1122), there are two stories associated with it that Bill told me and that had a significant impact on how I do science. I pass these along to all of my students.

First, Bill talks often about how Wodinsky insisted on taking notes in pencil. Bill thought this was a bit ridiculous, but he went along with Jerry. On his return trip home (New York), Bill took advantage of being able to buy very good gin at low prices in Bimini, and packed two bottles in his luggage along with the notebooks from the summer work. As one might guess, both bottles broke on the way home. But thanks to Jerry's insistence that the notes be taken in pencil (which is resistant to alcohol), the data were preserved even though the ink-drawn lines in the books

Fig. 25.1 Eugenie Clark and Bill Tavolga (about 2003). Dr. Clark, who is known internationally as the "Shark Lady," and Bill were graduate students together (along with Bill's late wife Margaret) at NYU, and both were students of Dr. Breder. I first met Genie in about 1967 when she was on the faculty of City College of New York. I later became Genie's "boss" when I served as Chair of Zoology at the University of Maryland and she was a professor in the department

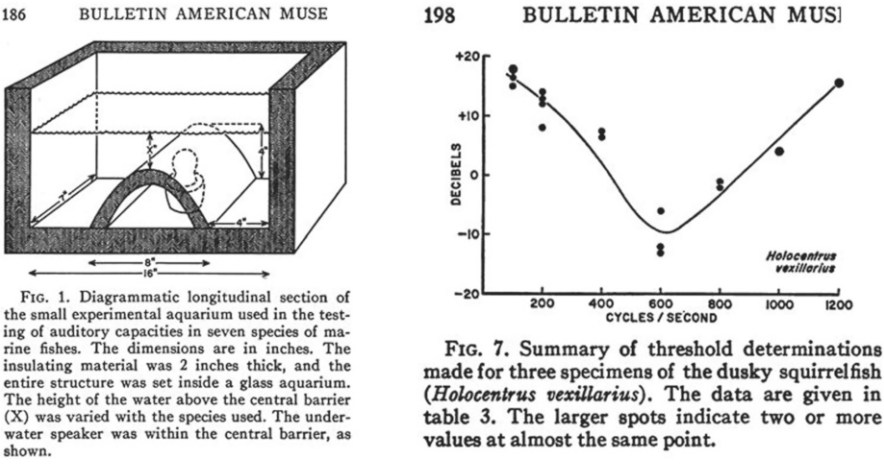

FIG. 1. Diagrammatic longitudinal section of the small experimental aquarium used in the testing of auditory capacities in seven species of marine fishes. The dimensions are in inches. The insulating material was 2 inches thick, and the entire structure was set inside a glass aquarium. The height of the water above the central barrier (X) was varied with the species used. The underwater speaker was within the central barrier, as shown.

FIG. 7. Summary of threshold determinations made for three specimens of the dusky squirrelfish (*Holocentrus vexillarius*). The data are given in table 3. The larger spots indicate two or more values at almost the same point.

Fig. 25.2 Figure from Tavolga and Wodinksy (1963). The left picture shows a shuttlebox used in the experiments. On the right is an audiogram they determined for the dusky squirrelfish. (Courtesy of The American Museum of Natural History)

were badly smudged. Our whole field would have been different had Jerry not insisted on pencil!

The second story Bill tells is about the actual study. The setup involved having a fish in a shuttlebox inside a chamber that was opaque on all sides but the top. A mirror was placed over the tank and reflected the view of the fish to another mirror across the room where the experimenter could monitor the response (this was before Bill and Jerry adopted photocells and 1960s modern electronics) and control the sound and the shock that would follow if the fish did not cross the barrier. Bill had trained a dusky squirrelfish (now *Sargocentron vexillarium*) to cross the barrier whenever it heard a sound and was testing its hearing by using the staircase (or tracking) method to raise and lower the sound level depending on the response of the fish in the previous trial (e.g., Fig. 25.2b). After some time, Bill found that the fish was detecting every sound played, no matter how low it was. Bill was quite befuddled until he realized that while he was watching the fish, the fish had learned to watch him and had figured out that when Bill moved his hand to turn on the sound, this would be followed by a shock. Thus, the fish learned not to respond to the sound per se but to the hand movement! The moral of this story is that one has to think very hard about the consequences of anything one does in experimental design and also work very hard to make sure that controls really are controls!

25.2 Blind Cave Fish and Georg von Békésy

I can trace my interest and excitement in biology to a single person—my sixth grade teacher, Thomas Vinci. Mr. Vinci was the only male teacher in my school (which went from kindergarten to sixth grade and had perhaps 40 teachers). He was (and is)

a truly gifted teacher. Although Mr. Vinci taught us many other things besides science, science was his first love, and he passed that love on to a number of his students, including me. Mr. Vinci encouraged science, fostered curiosity, and profoundly impacted the lives of his students.

Some years later, I was an undergraduate at the now-defunct Bronx campus (called the Heights) of New York University (NYU). On my walk to school one day I noticed a new pet store along the way. I walked in, looked at the tropical fishes, and "discovered" a tank with fish that did not have eyes, Mexican blind cave fish (then called *Anoptichthys jordani* but now *Astyanax mexicanus*). These fish totally intrigued me, and so when I got to campus, I went straight to my comparative anatomy professor, Douglas B. Webster (another amazing teacher), and asked him about these fish. Doug, being a consummate teacher, did not answer my question, but instead encouraged me to do research on the fish. This started as a library exercise but wound up in a two-year undergraduate research project on the general morphology of this species and its eyed ancestor. And because Doug's own research was on hearing and the ear (then working on desert rodents; e.g., Webster, 1962), I was also exposed to auditory neuroscience.

In learning about the fish, I read papers by the great ichthyologist Charles M. Breder, Jr. (e.g., Breder, 1943) and by one of his students Phyllis Cahn (Cahn, 1958). I mention this because both Drs. Breder and Cahn were on the faculty at the American Museum of Natural History (AMNH) in New York City, then, as now (in my view), the finest natural history museum in the world. Coincidentally, Doug Webster (who joined Dick Fay and me later to organize a meeting on the evolution of hearing; Webster et al., 1992) arranged for me to work at the AMNH for Dr. Donn E. Rosen (another student of Dr. Breder), chair of the Ichthyology Department. Here I met Phyllis Cahn, whose office was next to my work area.

Demonstrating serendipity in science, one day I was working on the skeleton of a large parrotfish (Family Scaridae) and in walked one of the other faculty in the Ichthyology Department, Dr. C. Lavett Smith. I cannot recall why Dr. Smith came in, but I do vividly recall his picking up the skeleton, taking out a structure he called an otolith, and telling me that it was part of the ear. While in Doug's lab, I was introduced to ears; I did not even know that fishes had ears, much less ear bones, until Dr. Smith came into my lab.

A few years later I was a graduate student at Queen's College of the City University of New York (CUNY) and looking around for a doctoral mentor. I heard about Dr. William Tavolga, a faculty member at another CUNY campus, City College of New York (CCNY). I suspect that one of the things that interested me about Tavolga was that he was one of the few among the faculty in CUNY working on fishes. It may also have been because Bill's lab was in the fabled and historic Department of Animal Behavior at the AMNH (the first animal behavior department in the United States) and I wanted to continue working at the AMNH.

On my first visit to Bill, he agreed that he might take me on as a student, but I had to come up with my own project. He gave me some papers and told me to come back in a week or so. I pondered the topic and then I found a comment in one of Bill's papers that said that no one yet knew if fishes can localize sound.

I suggested to Bill that I study sound localization, and his immediate response was that it would take 17 years to complete a study, and I signed on (although I did not believe it would take 17 years). Bill's point was that sound localization was one of the really difficult questions about fish hearing, and, in fact, Willem van Bergeijk (1964) had just argued very clearly that fishes cannot localize sound (see Chapter 7 by Fay and Chapter 14 by Hawkins for further discussions of fish sound source localization).

So how does one study something so enigmatic? I devised a plan to use the shuttlebox to train fishes to respond when they heard sounds shifting sides, with the idea that they would be able to localize a sound if they could detect shifts in the sound source. Then, Bill made the suggestion that I use a fish without eyes to avoid any visual cue and I came back to my blind cave fish. I won't continue the story with localization other than to say that Bill's 17 years were way off; even today, we really don't have a clear understanding of the mechanisms and capabilities of fishes for sound localization, although there is enough evidence to say that at least some species can do it (e.g., Fay & Popper, 2012). Because localization studies, at least in a lab on the fifth floor of the AMNH, were not possible (all kinds of issues with tank acoustics), I finally switched my dissertation to a study of comparative hearing in the Mexican blind cave fish and its eyed ancestors. This proved to be very tractable and was the first study to show, using psychophysics, that fish could hear to over 4 kHz (Popper, 1970). The study also showed that hearing in both the blind and eyed forms was about the same and that the two groups (they are now considered to be morphs of the same species) had similar hearing structures (Popper, 1971).

As an aside, I was immensely fortunate in meeting Bill Tavolga. He was (and is to this day) an amazing and caring mentor and exceptional scholar (and musician!). He not only facilitated my doctoral work, but he also provided a role model for mentorship and doing science for which I am deeply grateful. I also think it is fair to say that Bill "invented" the field of marine bioacoustics (Tavolga, 1964, 1967).

It also turns out that Dr. Breder, in whose work I read about blind cave fish, was doctoral advisor to Bill Tavolga (and Phyllis Cahn and Eugenie Clark; Fig. 25.1), and so Dr. Breder is my academic grandfather! I should also add that, quite independently, Bill had a major impact on my wife Helen as well. Helen met Bill in her first semester biology course at CCNY[2] when Bill was her lab instructor [CCNY had no teaching assistants in those days and so labs were run by professors] and then took other classes with him; Helen remembers Bill as an exceptionally good undergraduate instructor.

While doing my dissertation, I was invited by Phyllis Cahn, by then a major figure in lateral line research, to attend a 1966 meeting on lateral line in New York City (Cahn, 1967). The keynote speaker at the meeting was Georg von Békésy, winner of the 1960 Nobel Prize in Medicine and Physiology for his work on hearing (von

[2] Until the mid-1960s CCNY was strictly an undergraduate institution. It cost perhaps $50/year to attend, and was considered one of the finest colleges in the United States. Indeed, CCNY has produced more graduates who went on to win Nobel Prizes than any other college or university in the world –something like 10 or 11 to date.

Békésy, 1967). Von Békésy had just left Harvard and moved to the University of Hawai'i (where, unbeknownst to me then, I would move a few years later). I managed a few moments to speak with von Békésy and asked him what kind of research he would be doing in Hawai'i. His answer was that he was going to study fish hearing! I vividly recall a sudden feeling of depression when I realized that my whole field had disappeared; what could I contribute once this Nobel Prize winner got into the topic?

Several years later, when I was an assistant professor at the University of Hawai'i, I met von Békésy at the garbage dump of our apartment building; we both lived in faculty housing and he was (quite literally) our upstairs neighbor. During a later meeting, I asked him why he was no longer working on fish hearing, and his answer was that "it was too hard"! A feeling of depression again!

So, although von Békésy kept giving me doubts about my field, he also did me a great favor. For the first time in his career, von Békésy decided to invite a postdoc to join him, and he turned to his friend Professor E. G. Wever at Princeton. Wever sent a recent doctoral student of his to work with von Békésy; his name was (is) Richard Fay (see Chapter 7 by Fay). Dick and I met for the first time on December 26, 1971 when he, his wife Cathy, and their son Chris came to our house for a barbeque.

Indeed, in thinking back on it, although von Békésy never contributed to studies of fish hearing per se, perhaps his "arranging" for Dick and me to meet was his major contribution to the field. While I cannot begin to imagine how Dick's and my careers might have progressed separately, I think that we would both agree that as a team we've accomplished more than two individuals separately and we have had opportunities to contribute in ways that neither of us would ever have alone. Most certainly there would be no Springer Handbook of Auditory Research.

25.3 Fish Hearing

25.3.1 Early Comparative Studies

One of the most interesting questions with regard to fish hearing focuses on comparative issues, something that I was introduced to by Bill Tavolga. Indeed, one "difference" in approach for Dick Fay and myself is that I come from a strong comparative perspective, whereas Dick has focused on a wide range of studies of the hearing capabilities of one species, the goldfish (*Carassius auratus*). As a result of my comparative interests and the work done in a number of labs as well as my own, it is now clear that among the 32,000 or more species of fishes there are substantial differences in hearing capabilities and mechanisms (reviewed by Ladich & Fay, 2013).

The initial understanding of the variation in ear structure and the potential in capabilities came from the anatomical descriptions of Ernst H. Weber (1820), who described ears in a number of species and first described a series of bones, now known as the Weberian ossicles, that connect the swim bladder to the inner ear in the otophysan fishes (goldfish, catfishes, etc.; Fig. 25.3 next page). This was followed

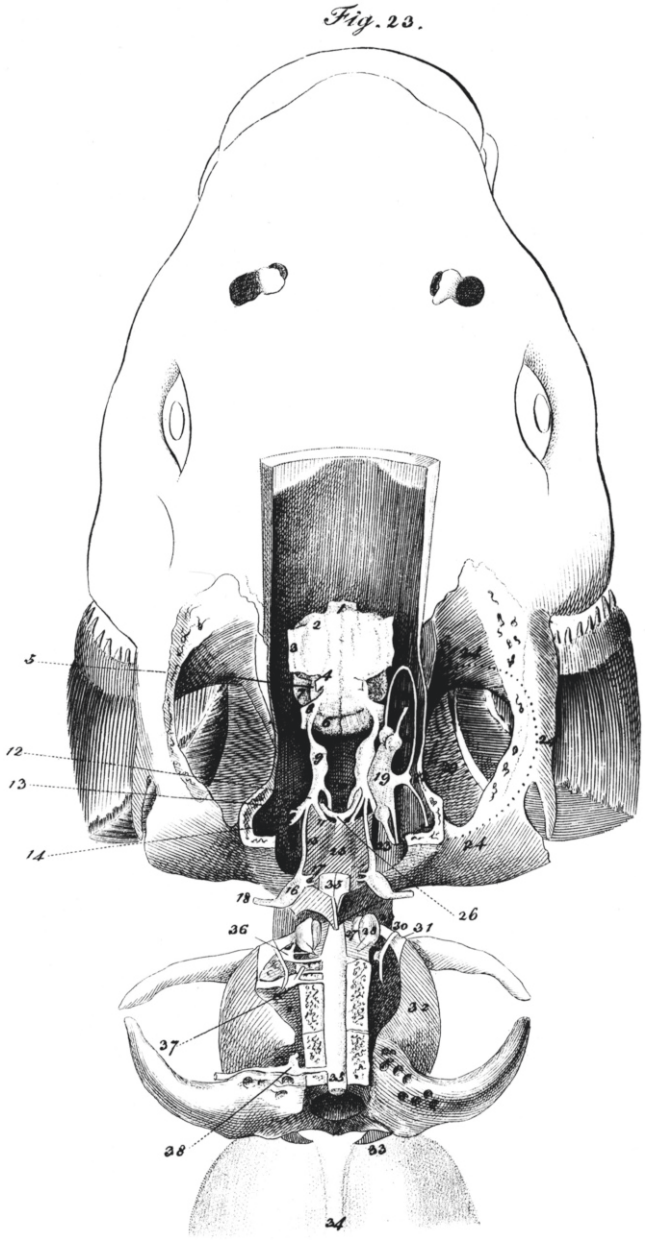

Fig. 25.3 (Above) Drawing of the head of a carp by Weber (1820, plate IV, figure 23) showing the head of a carp. The skull is opened in this picture and the brain cut away to show the ears. The right ear is labeled no. 19. The Weberian ossicles are the bones to the left and right of the vertebral column just behind the skull. (Next Page) Drawings of the ears of the salmon (*Salmo*) from Retzius (1881, plate XIV). I thank David Corey for providing the very high resolution image from Retzius shown here

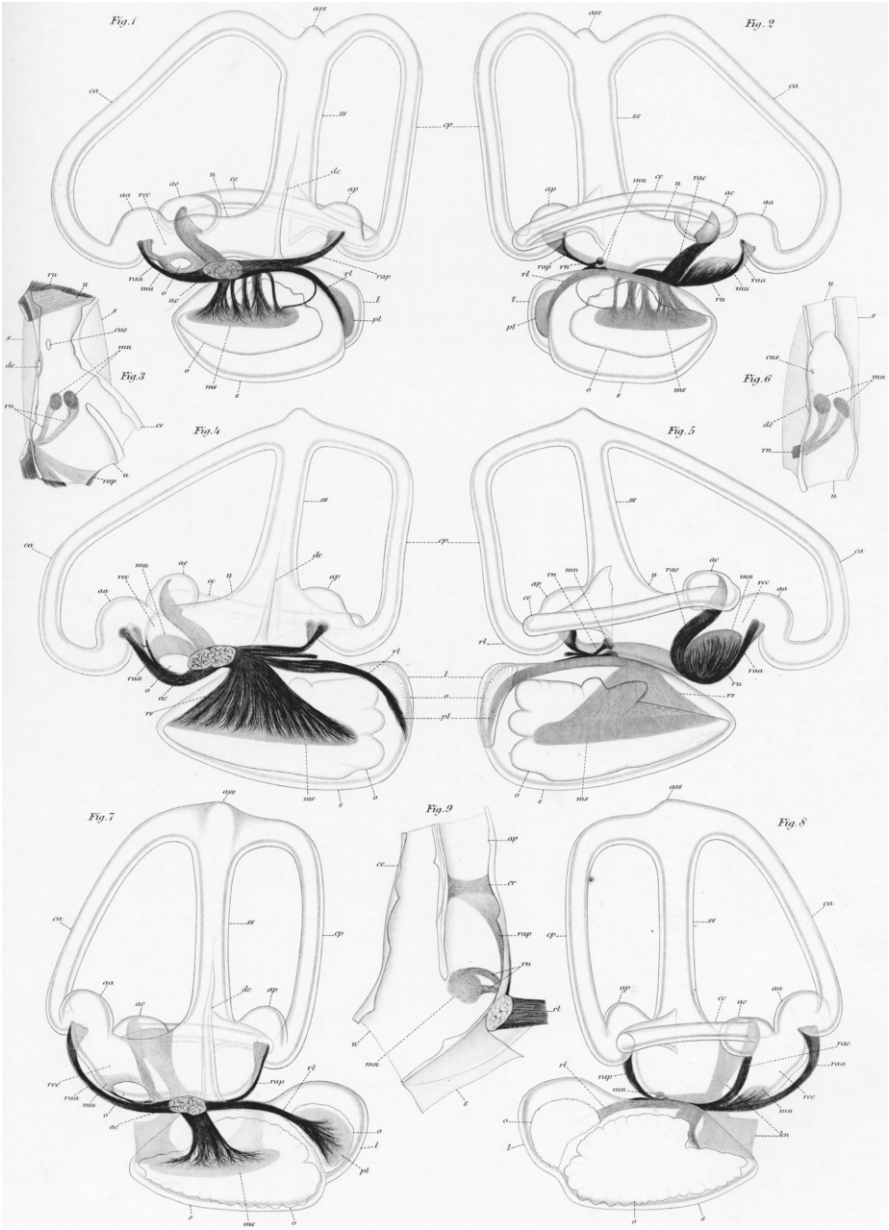

Fig. 25.3 (continued)

by Gustav Retzius (1881), who described the ears, and its variations, in dozens of fish species (Fig. 25.3 above). Indeed, Retzius' anatomical drawings are still invaluable today in helping us understand the variations in the ears of fishes and they have been

instrumental in helping decide which species to study for measures of hearing capabilities. One conclusion coming from the body of work by Retzius is that there is probably far more diversity in ear structure in fishes than in all the other vertebrates combined.

25.3.2 Comparative Hearing

Fish hearing capabilities vary in several dimensions. Most notably, different species have different hearing bandwidths. The narrowest hearing ranges, from below 50 Hz to perhaps 500 Hz, are found in species that do not have a swim bladder (an air chamber in the abdominal cavity that likely evolved for buoyancy control but that later evolved for use in hearing and sound production in many species). Other species may hear up to 1000–1500 Hz, and these often involve the swim bladder to a greater or lesser degree (reviewed in Popper et al., 2003).

A third "group" of fishes hears sounds up to 3000–4000 Hz. These fishes have a specialized connection between the inner ear and the swim bladder that enhances the hearing range and hearing sensitivity (Jacobs & Tavolga, 1967; Popper, 1971). The goldfish, for example, has a series of bones, the aforementioned Weberian ossicles (Fig. 25.3), that serve as a direct path for sound from the swim bladder to the inner ear, whereas some squirrelfish (genus *Myripristis*) have anterior projections from the swim bladder that directly contact the inner ear (Coombs & Popper, 1979).

We now have hearing data on more than 100 species of fish (see Ladich & Fay, 2013), and what is striking is the wide variation in hearing bandwidth and thresholds for various species. However, as Fritz Ladich and Dick Fay (2013) point out, much of these data have to be considered with care because thresholds were often measured in terms of sound pressure and not in terms of particle motion, the major auditory stimulus in most species (e.g., Popper & Fay, 2011; see also Chapter 14 by Hawkins). Moreover, with few exceptions, most studies of hearing have been done in small tanks where, as my late friend Antares Parvulescu (1964) clearly pointed out, it is virtually impossible to calibrate the sound field. As a consequence, though many of the studies to date are important and reveal a good deal about interspecific variation in fish hearing, we are still a reasonably long way from knowing as much as we need to know about auditory sensitivity in fishes, and particularly as it relates to detection and use of both pressure and particle motion.

25.4 Comparative Ears

There is extraordinary diversity in the structure of fish ears. As noted by my close friend Christopher Platt, from the great lithographs of Retzius on vertebrate ears (Retzius, 1881), much variation is found in the semicircular canals (Platt, 1983). Though some of this variation may be related to the shape of the skull bones in which

the canals are embedded, some variation in these tubular shapes may determine sensitivity to certain frequencies or amplitudes of head movements (see Platt, 1983).

The variation that has most intrigued me is in the three otolithic end organs: the saccule, lagena, and utricle. Although it was first proposed by von Frisch (1923) that the saccule, and perhaps the lagena, are the main hearing organs in fishes, recent evidence strongly supports the idea that the utricle may be involved in hearing as well (and see Section 5 on ultrasonic hearing).

In 1975 I had the opportunity to learn scanning electron microscopy (SEM) and apply it to fish ears. I first examined the saccular epithelium of the lake whitefish (*Coregonus clupeaformis*) and recall one of the "eureka" moments in my career, the realization that rather than having hair cells oriented in two opposing directions as had been described for virtually all other vertebrates analyzed to date (e.g., Wersäll et al., 1965), this salmon relative had saccular hair cells oriented in four directions, two dorsoventrally (as in all other saccules) and two rostrocaudally, something that had never been observed (Fig. 25.4). This work was published in *Science* (Popper, 1976). To be fair, just as my paper was published, another paper came out in Europe by Tor Dale (1976) on the ear of the Atlantic cod (*Gadus morhua*) that also showed hair cells oriented in four directions, confirming that this "plan" is not unique to one fish group.

Following this study, I was involved in a series of investigations that explored the structure and ultrastructure of the ears in widely diverse species (e.g., Popper, 1977, 1978, 1980; Popper & Platt, 1979; Popper & Northcutt, 1983). These investigations revealed that there is substantial variation in the hair cell orientation patterns in fishes, with particular focus on the saccule. We found that most fishes have saccular hair cells oriented in at least four distinct directions (Figs. 25.4 and 25.5). However, the plan was often not nearly as "simple" as found in lake whitefish or Atlantic cod, but instead, the orientation patterns on the rostral end of the epithelium were often complex and highly specialized (Figs. 25.5 and 25.6).

Several questions arose. First, why do fishes have variation in saccular hair cells (with there being much less variation in lagenar hair cells and even less in the utricular hair cells; see Section 5)? Second, why are their hair cells oriented in multiple directions? Both questions are still open, but we think we have some basic suggestions for both.

25.4.1 Variation

For her doctoral dissertation in my lab, Sheryl Coombs examined hearing in several different species of squirrelfish (Holocentridae). She found that a species of *Adioryx* could hear to about 1500 Hz, whereas the closely related species *Myripristis* was able to detect sounds up to 4000 Hz (Fig. 25.5) (Coombs & Popper, 1979). Putting this together with work from Tavolga and Wodinsky (1963) on a third species (Fig. 25.2), we came to the conclusion that perhaps the variation in hearing could be correlated with hearing structures. This idea was supported by a morphological

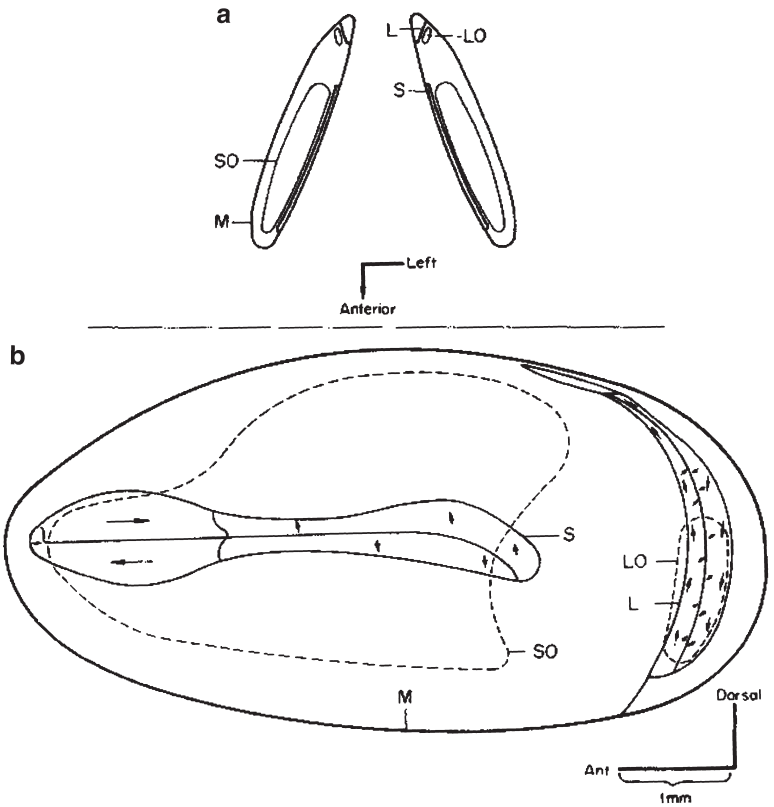

Fig. 25.4 Hair cell orientation patterns from lake whitefish (Popper, 1976). (**a**) Dorsal view of the two ears showing the lagena (L) and its otolith (LO) and the saccule (S) and its otolith (SO). The otolith lies in close proximity to the sensory epithelia in each end organ. (**b**) A lateral view of the saccule and lagena showing the otoliths (dashed lines) and the sensory epithelia. The hair cells are divided into "orientation groups" based on the position of the kinocilium in each ciliary bundle relative to the rest of the bundle. The tip of the arrow indicates the direction of orientation (toward the kinocilium) in each region. Regions are divided by solid lines

study by O'Connell (1955), who showed that the swim bladder is farthest from the ear in *Adioryx*, progressively closer in *Holocentrus*, and intimate to the ear in *Myripristis*. Moreover, my studies of the saccular epithelium in the two species that Sheryl studied showed that the epithelium in *Myripristis* is far larger and more complex in terms of orientation patterns than that in *Adioryx* (Fig. 25.5) (Popper, 1977).

Sheryl and I then started to examine what we knew about hair cell orientation patterns and hearing in a wide range of species. We proposed the hypothesis that fishes with the most highly elaborate (in terms of orientation pattern) saccules inevitably are species that (a) have a wider bandwidth of hearing and (b) have specializations that somehow mechanically "connect" the swim bladder to the inner ear (Popper & Coombs, 1982). Indeed, we then predicted that we could make

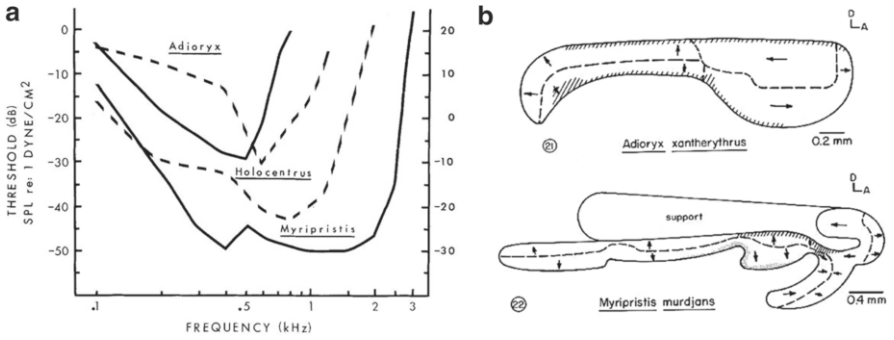

Fig. 25.5 Hearing capabilities and hair cell orientation patterns in squirrelfish. (**a**) Hearing thresholds for various species of squirrelfish. Solid lines are for a species of *Adioryx* and *Myripristis* measured by Coombs and Popper (1979). The dashed lines are for another *Adioryx* species and a species in the genus *Holocentrus* as determined by Tavolga and Wodinsky (1963). (Figure from Coombs and Popper, 1979.) (**b**) Saccular hair cell orientation patterns for the species used by Coombs and Popper (from Popper, 1977). The figure illustrates that the species with the most elaborate hair cell orientation pattern also has the widest hearing bandwidth. Interestingly, *Holocentrus* has hearing capabilities between those of the other two species, and there is evidence from O'Connell (1955) that the adaptation for enhanced hearing lies between the two other species. We do not, however, have ultrastructural data for any species of *Holocentrus*

suggestions about the bandwidth of hearing in fish just from seeing the ultrastructure and hair cell orientation pattern on the saccule.

Since then, we have tested this idea a number of times. For example, John Ramcharitar, when a graduate student in my lab, examined the ears and hearing in a number of species of Sciaenidae (croakers), a commercially very important group of fish (e.g., Ramcharitar et al., 2006). John found a very close correlation between hearing bandwidth and complexity of saccular structure, with fishes having the most complex structure having the widest bandwidth.

To my thinking, the ultimate test for this hypothesis would come from work that I did in the 1980s on the ears of species from very great depths (several thousand meters) and from work recently completed by my last graduate student Xiaohong Deng and an undergraduate working with Xiaohong, Bradley Buran (Popper, 1980; Buran et al., 2005; Deng et al., 2011, 2013). In these studies, we found that most deep-sea species we examined have highly specialized saccules (e.g., Fig. 25.6a), often with hair cells having exceptionally long ciliary bundles. Moreover, Xiaohong has shown that similarly distinct orientation patterns and other ultrastructure features show up time and again in taxonomically unrelated species. Thus, although we predict that many deep-sea species (living in areas without light) are likely to have excellent hearing based on the structure of their ears, it is virtually impossible to do hearing tests on them because they cannot be kept alive when brought to the surface.

At the same time, I don't want to leave the impression that fishes must have four hair cell orientation patterns in the saccule to detect higher frequencies (3000–4000 Hz). For example, goldfish are known to hear to 3000 Hz (Jacobs & Tavolga, 1967; see Chapter 7 by Fay), yet have a relatively simple bidirectional saccular

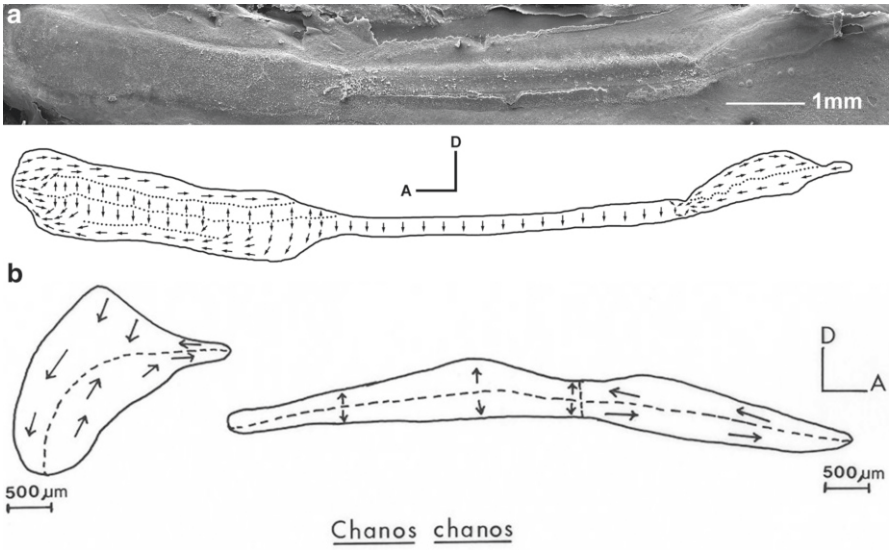

Fig. 25.6 (**a**) Scanning electron microscopic view (top) and drawing of hair cell orientation patters on the saccule of the deep-sea fish *Antimora rostrata* (blue antimora). This species, typical of many other deep-sea species, has a very complex hair cell orientation patter on the rostral end of the epithelium (left). In addition, there are rostral–caudal oriented cells and the caudal end of the epithelium as well, something found in some, but far from all, taxonomically diverse species. (Image from Deng et al., 2011. *Deep-Sea Research I*. Reprinted with permission). (**b**) Hair cell orientation patterns from the saccule (right) and lagena (left) of the milkfish *Chanos chanos*. Note that the rostral end of the saccule (right) has hair cells oriented rostral-caudally, whereas its descendants, the otophysan fishes, have an equally elongate saccular epithelium but only hair cells oriented dorsally and ventrally (like the caudal end of the milkfish saccule). (From Popper and Platt, 1983, *Journal of Morphology*, *reprinted with permission*)

pattern. Platt (1977) did an SEM analysis of the hair cell orientation patterns in all the end organs in the ears of goldfish (and later zebrafish [*Danio rerio*]; Platt, 1993) and showed that the whole saccule has only dorsally and ventrally oriented hair cells. This pattern has been confirmed for all members of the Otophysi (fishes with Weberian ossicles) (Platt & Popper, 1984). The ability to hear well in the Otophysi is no doubt related to the presence of the aforementioned the Weberian ossicles. It may be that their relatively simple saccular pattern is related to oscillation vectors produced by this specialized direct coupling of the gas bladder to the sensory macula. In any case, this finding of Platt and others argues against hearing specialization always being correlated with four hair cell orientation patterns.

However, Platt and I obtained samples of the milkfish *Chanos chanos*, a species representing the ancestor of all of the otophysan fishes. We discovered that *Chanos* has a saccule that is intermediate between the modern Otophysi and fishes with four hair cell patterns; the shapes of the saccule and lagena are very similar to those in Otophysi, but the saccule has hair cells oriented in four directions (Fig. 25.6b) (Popper & Platt, 1983). *Chanos* also has bones that are clearly on the way to becoming Weberian ossicles. The conclusion we reached was that the presence of only two

hair cell orientation directions in the Otophysi is a derived characteristic and one that is an extreme specialization for a broad bandwidth of underwater hearing!

In fact, this idea is supported by work done by Cathy McCormick when she was a postdoc with me. Cathy studied hearing in mormyrid fishes (*Gnathonemus* sp.), elephant-nosed fishes that use electroreception for communication. She found that *Gnathonemus* has hearing sensitivity that rivals that of the Otophysi (McCormick & Popper, 1984), although they are not at all closely related taxonomically, and we found that the saccular hair cell orientation pattern is bidirectional (Popper, 1981) as in Otophysi. However, instead of having Weberian ossicles, there is a large air bubble attached to the saccule in this species, thereby enhancing hearing capabilities.

Indeed, having bidirectional hair cells in the saccule appears to be the "norm" for other vertebrates, whereas four directions are found in many bony fishes. This idea is supported by findings that show that sharks and rays have saccular hair cells oriented in two directions (Corwin, 1981a), and we have found the same thing in a whole range of more primitive fish that representing the origins of the species that have hair cells in four directions (Popper, 1978; Popper & Northcutt, 1983; Mathiesen & Popper, 1987). Moreover, a similar pattern is found in lungfishes, a group that is thought to have given rise to terrestrial vertebrates (Platt et al., 2004).

25.4.2 Why Multiple Hair Cell Patterns?

Of course, a singularly interesting question is why so many fish species have hair cells oriented in multiple directions. Related to this is the question of why fishes with particularly wide hearing bandwidths often have even more elaborate orientation patterns. And, although not being discussed here, what is the functional significance for fishes of having, on a single epithelium, hair cells with different length ciliary bundles (Popper, 1977; Platt, 1983; Popper & Platt, 1983)?

One very reasonable suggestion for having hair cells oriented in different directions may reflect back to the question I initially wanted to ask for my dissertation, sound source localization. Fishes do not have available to them the same acoustic cues for localization used by terrestrial vertebrates (e.g., interaural differences) owing to the close proximity of fish ears to one another and the much higher speed of sound in water than in air (e.g., van Bergeijk, 1964). At the same time, the basic mechanism by which the sensory hair cells of the ear are stimulated results from the relative motion between the dense overlying otolith and the sensory cells, and this motion, which is in response to the particle motion component of the underwater sound field, is directional (Popper et al., 2003; Fay & Popper, 2012).

Thus, one can imagine that if the relative motion between the epithelium and otolith changes with the direction of the impinging sound field, and knowing that the physiological response of the hair cells is directional, it follows that by having hair cells oriented in different directions, each would respond maximally to sound from different directions. We therefore speculated that by combining inputs from hair cells oriented in multiple directions, it should be possible to localize a sound source (Rogers et al., 1988). Although the actual process is somewhat more

complex owing to the presence of pressure as well as particle motion fields and many other factors (e.g., Rogers & Zeddies, 2008), it is likely that one important role of hair cells oriented in different directions is to aid in determining the direction of a sound source. At the same time, when one considers that each of the three end organs in most fishes lie on different planes, this means that not only can fishes use directional responses from the two saccules, but also that the other end organs can potentially contribute to directional responses as well, thereby refining the information about direction.

Still, sound localization by fishes remains an enigmatic problem. Although it is clear that the hair cell orientation patterns of the ear are involved in determining direction, there are few data showing how well fishes can localize (e.g., Rogers & Zeddies, 2008). This remains an area that, even 40+ years since my doctorate, has not been solved, making that Tavolga's 17-year prediction way off!

25.4.3 So Why Variation?

But the question still remains as to why some fishes have more complex orientation patterns than others and why many of the species with the most complex patterns have a wider bandwidth of hearing than fishes without complex patterns. Perhaps this has to do with refinements in systems for sound localization. Or perhaps this has to do with other aspects of hearing.

There are two basic hypotheses (Popper et al., 2003). One is that in the evolution of numerous species, fishes have "experimented" in widely different ways to extract the same information from sound. That is, as we pointed out years ago (Fay & Popper, 2000), for fishes to glean the maximum amount of information from the acoustic scene, they need to be able to detect sound, discriminate between sounds, localize sound, and detect signals in the presence of noise. If we assume that every fish needs to be able to do these functions, then it is possible that the different inner ear patterns are all different ways to help accomplish the same tasks (also see Chapter 7 by Fay). Alternatively, if all fishes do not have to do the same basic things in terms of hearing, it is possible that the different patterns have evolved to do different auditory tasks. There is no ready way to resolve which hypothesis is correct, but this is a question worth asking in the future.

25.5 Ultrasound Detection

In the late 1990s, I read several papers that suggested that some fishes in the herring family (Clupeidae) could be kept from entering the water intakes of nuclear power plants by projecting ultrasound into the water around the intakes (e.g., Dunning et al., 1992). This was my first introduction to the idea that sound could potentially be used to control fish behavior, and I found the whole idea of fish detecting ultrasound somewhat "ridiculous." But, because clupeids are some of the most commercially important fishes in the world, we submitted a proposal to the National Science

Foundation to examine hearing, and ultrasonic hearing, in a clupeid fish, the American shad (*Alosa sapidissima*). The grant was funded (those were the days when "wild" ideas could still get funding) and David Mann, then a postdoc in the lab, led a project to explore hearing in this species. Another postdoc (and former Fay doctoral student) Zhongmin Lu collaborated with David on the project.

We discovered that American shad and their relatives in the subfamily Alosinae (shads, menhaden, and some freshwater herring) are able to detect sounds to at least 180 kHz (Mann et al., 1997). Thus, these fishes may have the widest hearing bandwidth of any known vertebrate including echolocating dolphins and bats.

Once we realized that Alosids detect ultrasound, we struggled to figure out why they have such an extraordinary hearing range. We finally realized that perhaps these fishes are detecting the high-frequency echolocation sounds of dolphins and avoiding predation. Although the idea seems far-fetched, it, in part, arose from our familiarity with the work on moths and other insects that have evolved high-frequency hearing to detect echolocating bats and avoid being eaten (e.g., Roeder & Treat, 1961).

Although it was not possible for us to expose American shad to actual dolphins, my postdoc Dennis Plachta developed a behavioral paradigm that exposed American shad to ultrasonic signals and enabled us to observe the response (Plachta & Popper, 2003). We found that the American shad did not show a behavioral response to low-frequency sounds (e.g., 500 or 1000 Hz) but that they would react to ultrasonic signals. We also found that responses of American shad to ultrasound, like those in moths evading bats, were "graded." At lower received intensity signals, the American shad would swim away from the source (demonstrating, by the way, sound source localization). As sound levels got higher, the responses became more rapid, and at the highest sound levels, the fish showed highly random and "chaotic" behavior. The conclusion we reached is that when an echolocation click is just audible, the American shad may not pay attention, "thinking" that the dolphin was far away. But as the sound gets louder, and potentially the dolphin closer, the fish start to actively swim away from the sound source. Finally, if the dolphin is very close and the sound loud, the fish school (often tens of thousands of fish) go into random rapid motion so that the predator can no longer echolocate on a single animal, thereby lowering the likelihood of predation.

Along with trying to understand why American shad detect ultrasound, it is equally of interest to know *how* ultrasound detection operates in a fish. This turned out to be a far more difficult problem and one that has yet to be solved.

The mechanism we proposed for ultrasound hearing involves use of the very highly specialized utricle that is known in clupeids to be in close contact with a small gas bubble in the head (Denton & Gray, 1979). This utricle, unlike those found in any other species of vertebrate, is tripartite (Popper & Platt, 1979). Moreover, the central epithelial region in ultrasound-detecting species is very thin, as demonstrated in a project led by my postdoc Dennis Higgs (Higgs et al., 2004). Thus, we hypothesized that the bubble resonates at ultrasonic frequencies and stimulates the middle utricular epithelium.

The problem in testing this hypothesis is that all clupeids are very fragile, with the lab "joke" being that just looking at a clupeid can result in its dying. Although American shad are perhaps the heartiest of the clupeids, even these fish could not tolerate

electrodes being placed near the utricle because the only possible surgical approach would be through the air bubble by the utricle. This would destroy the bubble and, of course, eliminate its being tested for ultrasonic hearing. However, Plachta, working with postdoc Michele Halvorsen and our colleague Jiakun Song, devised an approach that allowed us to test ultrasonic responses at various brain levels (Plachta et al., 2004). These studies showed that there are, indeed, neurons in the brain that respond best to ultrasound and there was good evidence that these units arose in the utricle.

Of course, things may not be as simple as we thought. Recent work by Wilson et al. (2009) suggests that connections between the air chamber in the head and the lateral line receptors on the body may also play a role in ultrasound detection. Although we are still convinced that the ultimate receptor is the utricle and that the air bubble is involved, the whole mechanism may be rather more complex and something that one would hope would be studied by future investigators.

Another question that arose was *how* fishes could have evolved ultrasonic hearing. Although there is no fossil record to give an answer, we do think we have a reasonable answer. The answer comes out of a paper by two friends and colleagues, Peter Rogers and Mardi Hastings (Rogers & Cox, 1988).

Rogers and Cox pointed out to that sound propagation is very different in shallow versus deep water. In shallow water (e.g., streams, shallow rivers), low frequencies propagate very poorly, and only higher frequencies propagate greater distances—the shallower the water the poorer the propagation of low frequencies. We thus proposed that fishes that have evolved sound detection above 1 kHz or so probably arose in shallow water, and, indeed, most fishes that hear above about 1500 Hz are in shallow water or evolved from species there. (As an aside, the same observation was made by Pliny the Elder [1890, p. 547], who noted that "…the mullet, the wolf-fish, the salpa, and the chromis, have very exquisite sense of hearing, and that it is for this reason that they frequent shallow water.") We therefore proposed that the ancestors of clupeid fishes evolved in shallow water. And, indeed, American shad and many other clupeid species are anadromous; they breed in shallow rivers and streams and then move out to the ocean to grow, as described by the Pulitzer Prize winning author John McPhee (2003).

Exactly why American shad and other *Alosa* evolved ultrasound detection is not clear, but perhaps selective pressures placed on them by echolocating dolphins resulted in the increased bandwidth, using the same utricular structures that are in all other clupeids. Again, this is a really interesting problem for future study.

25.6 Addition of Sensory Hair Cells in the Ear

In the early 1980s, Jeff Corwin (who completed his MS with Albert Tester, Ian Cooke, and me in Hawai'i) did a series of studies showing that sharks and rays had very large numbers of sensory hair cells in their ears and also that there was a continuous addition of such cells over the life of these animals (Corwin, 1981b). We started to wonder if the same phenomenon occurs in bony fishes. Thus, Becky Hoxter and I examined the number of sensory cells in the ears of different-sized

Oscars (*Astronotus oscellatus*) and found a substantial proliferation of cells in the saccule as the fishes grew (Popper & Hoxter, 1984). This has been confirmed for other species but perhaps most dramatically in a study done by my postdoc Antoni Lombarte. Toni, a fisheries biologist from Barcelona, obtained specimens of the Mediterranean hake (*Merluccius merluccius*), a relative of the Atlantic cod. We demonstrated a very substantial addition of sensory hair cells in each of the otolithic end organs for at least the first nine years of life and that this increase was several hundred cells a day (Lombarte & Popper, 1994). Nine-year-old animals had more than 2 million hair cells in each ear. Although speculative, we also noted that the largest "spurt" of hair cell addition came at about an age when *Merluccius* switched its feeding habits and started to feed on myctophid fishes, one of the most widely distributed fish groups and one thought to make sounds. Our very tentative suggestion was that *Merluccius* start to hear their prey when they switch to myctophids as a diet, and so hearing becomes more important in their lives.

Still, there is nothing known about why fishes increase hair cells. One possibility is that as fishes add cells, they increase hearing sensitivity. However, this makes little sense because it would mean that large fishes would have different hearing capabilities than smaller brethren, affecting communication and the detection of the acoustic scene. Alternatively, because fishes continue to grow through most of their lives, the structures associated with hearing change relative positions and the additional sensory cells are needed to maintain hearing at a certain level. Two lines of evidence support the latter hypothesis. First, in a study of walleye pollock (*Theragra chalcogramma*), we showed that the hearing sensitivity of small fish to both pressure and particle motion was virtually the same as that in fish 3 years older (Mann et al., 2009). Second, in a study on zebrafish hearing that he did while a postdoc, Dennis Higgs showed that hair cell proliferation stopped when zebrafish (which do not seem to grow substantially after they reach a certain adult size) stopped growing (Higgs et al., 2002).

Although there is no space to go into it here, one other point about the ability of fishes to proliferate new hair cells for most (if not all) of their lives is worth noting. In the early 1990s my postdoc Hong Young Yan demonstrated that treatment with the ototoxic drug gentamicin would destroy sensory cells in some parts of each sensory epithelium (Yan et al., 1991). This was followed by a study led by Lombarte, who showed regeneration of the cells over about 10–15 days post exposure to gentamicin (Lombarte et al., 1993). More recently, my postdoc Michael Smith and graduate student Allison Coffin, studying temporary threshold shift (TTS) in fishes, showed that exposure to loud sounds results in damage to sensory cells as well as TTS in some species, and that hearing recovers as hair cells return (Smith et al., 2006).

25.7 Bridge Construction and Other Applied Issues

As mentioned in Section 5, the study of ultrasound detection arose because of the use of high-frequency sounds to control fish behavior. Before hearing about this, I had never given thought to an applied use for our work on fish hearing, but over

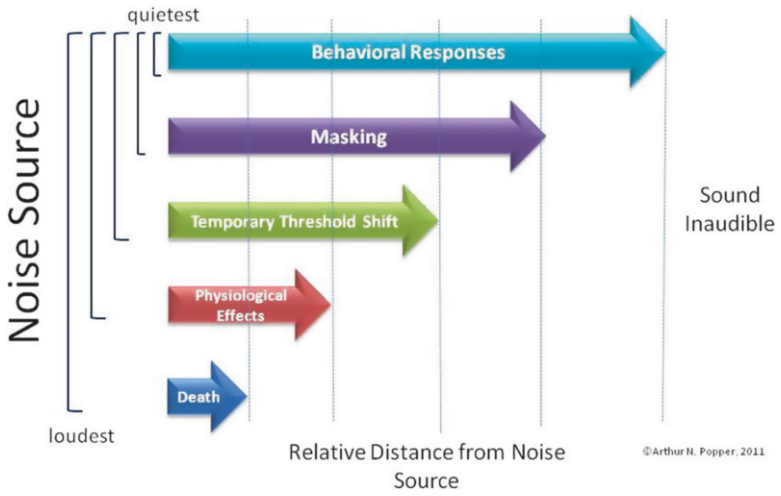

Fig. 25.7 Relationship between noise levels, relative distance, and potential effects

the past 10 years or so, the work in my lab has been heavily focused on asking questions that directly relate to the use of sound to control fish behavior and, far more importantly, on the effects of man-made sound (also called anthropogenic sound) on fishes. Indeed, Dick Fay and I looked back at a paper we published 20 years earlier (Popper & Fay, 1993; Fay & Popper, 2012) and came to the conclusion that one of the major driving forces now and for the foreseeable future for studies of fish hearing lies with these, and related, more applied questions.

The issue of effects of man-made sound on fishes is worldwide. Human activities are increasingly adding sound to the aquatic environment from a variety of sources. Commercial ships (e.g., oil tankers) are very noisy and, along with other kinds of boating, tend to increase ambient noise levels (Fig. 25.7). This may be particularly the case in places like shipping lanes and harbors, where increased ambient noise has the potential to mask sounds that are of biological relevance to fishes, including their own communication sounds and sound of the acoustic scene.

Perhaps more dramatic, however, are sounds produced in the construction of structures such as bridges and wind farms and from geological exploration for undersea gas and oil. These sounds tend to be very intense (in some cases over 220 dB re 1 μPa rms) and have the potential to kill or dramatically affect the behavior of fishes.

One of the real problems in considering the effect of man-made sounds on fishes is that the number of well-controlled and peer-reviewed studies is very limited (Popper & Hastings, 2009). Investigations of the effects of intense sounds on fishes are very difficult because the sound sources of concern cannot easily be brought into a lab because of their sizes. And studies in the field are very difficult because the

sound-producing devices used in construction and/or exploration are very expensive. As a consequence, investigators have no control over the sources during their experiments.

For example, one of the sources of concern is the seismic air guns used in oil and gas exploration. The specialized vessels and sources used cost hundreds of thousands of dollars a day to operate, so no vessel operator will "donate" time for a research project. Similarly, the major source of sounds during construction is from impact driving of piles used to support structures such as bridges and wind turbines. Pile drivers are very large machines (and very expensive to operate) and so they cannot be brought into the lab or purchased for use in an experiment.

Despite these limitations we have been able to make a number of contributions over the past few years that not only help to understand the effects of these intense sources on fishes, but, perhaps more importantly, to contribute directly to developing regulatory practices that both protect animals and allow needed construction and exploration.

25.7.1 Seismic Air Guns in the Arctic Circle

There is major international concern over the potential effects of seismic air guns on fishes (see references in Popper & Hawkins, 2012). These devices use release of highly compressed air to project sounds into the substrate. The reflected signals are picked up by long arrays of hydrophones and the data are analyzed to determine geologic formations that indicate the presence of gas and oil. In 2004 David Mann and I were invited to work with Bruce Hanna and Peter Cott of the Canadian Department of Fisheries and Oceans in Inuvik, Canada, to determine if the sounds from a small air gun array used in the Mackenzie River (the second longest river in North America) was harming local fishes. This was a great concern of the local Inuit population because they are subsistence fishers.

We exposed several species to air guns and measured hearing post exposure to determine if there was hearing loss and any damage to the sensory cells of the ear. This was the first study to measure hearing in fishes exposed to air guns. (Earlier, working with Rob McCauley in Perth, Australia, we showed that exposure to a seismic air gun could damage sensory hair cells in the ears of one species of fish; McCauley et al., 2003.) The fish were exposed to sounds up to received sound levels of around 198 dB re 1 μPa rms and then tested to ascertain whether there was TTS. We found that several of the species showed TTS, but that complete recovery took place within about 18 hours. Exposure to this intense sound did not kill any animals nor was there any evidence of damage to internal tissues (Popper et al., 2005). Subsequent analysis of the sensory epithelia of the inner ear also showed no effect (Song et al., 2008). This study was the first to examine potential hearing loss in any fish exposed to very high-intensity impulsive sounds, and it succeeded in showing that fish exposed to high levels of such sounds may not be killed or permanently harmed and that not every species shows the same effects from such exposure as others.

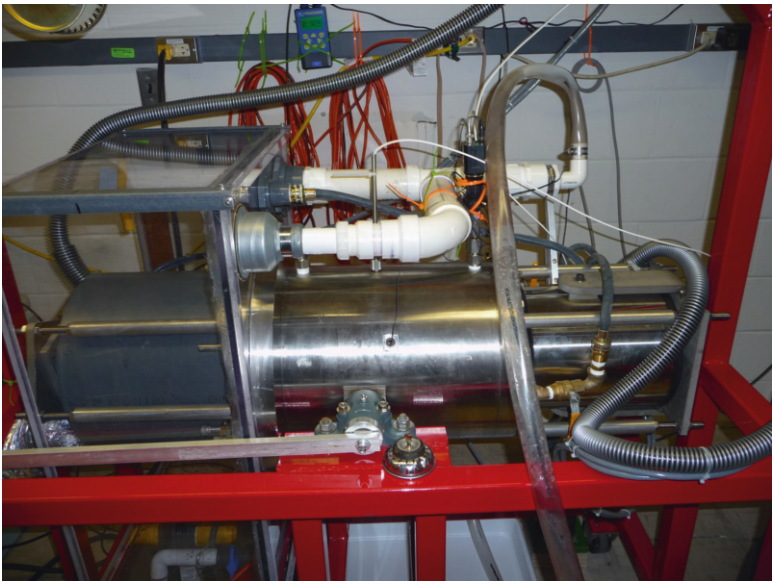

Fig. 25.8 A picture of the HICI-FT used in studies of effects of pile driving on fish. The large gray chamber on the left and the steel cylinder on the right are the shakers used to produce the sounds (the one on the left looks different because it is enclosed in plastic to keep out water when fish are put into the device. The central steel region is a "fish tank" in which the animals are exposed to sound. See Halvorsen et al. (2012b) for a description of how the device works. The hoses and plastic tubes are for temperature control and water flow

25.7.2 Pile Driving

During construction of the east span of the San Francisco Bay Bridge in the mid-2000s, concern arose that the very extensive pile driving may kill or harm migrating endangered salmon that pass the construction site. There was then very little useful scientific data to actually help understand the effects of the pile-driving sounds on fishes since it was not easy to take a pile-driving apparatus into the lab to do controlled studies.

One morning over breakfast, however, my colleague Tom Carlson and I came up with an idea to bring pile-driving sounds into the lab with the same sound levels and acoustic characteristics as found during actual pile driving in the field. The idea involved use of a device invented by Pete Rogers, something he called a "ratabrator." Tom and I managed to get funding and then worked with Pete and his group to redesign the "ratabrator" so that it could generate very high intensity pile-driving signals in my lab at the University of Maryland. The device was named the HICI-FT (pronounced hissy fit) (Fig. 25.8) by my postdoc (and project leader) Michele Halvorsen because of its initial fickleness. Once we figured out how to isolate the HICI-FT from the building structure (its first uses shook our five-story steel-framed building!) and cool it so that the very powerful shakers needed to generate the sound field would not heat the water and cook the fish, we were able to do a range of experiments on the effects of pile-driving sounds on fish.

In initial studies on young Chinook salmon (*Oncorhynchus tshawytscha*), we showed that the onset of physiological effects (damage) occurred only when fish were exposed to accumulated sound energy (cumulative sound exposure level [SEL_{cum}]) of 210 dB re 1 $\mu Pa^2 \cdot s$, a signal level that was equivalent to giving the fish 1920 pile strikes (at 1.2-s intervals) of 177 dB re 1 $\mu Pa^2 \cdot s$ total sound energy in each strike (Halvorsen et al., 2012b). These provide the experimentally derived sound exposure levels that can be used in developing regulations to protect the fish. At the same time, these proposed levels are substantially higher than those currently used by regulators and that are not science based, meaning that although protective of fish, the levels from our studies also mean that construction is less likely to harm the animals than previously thought.

Subsequently, with the added collaboration from my last postdoc Brandon Casper, additional studies have shown that the levels that result in the onset of physiological effects are about the same for a morphologically diverse group of species (Halvorsen et al., 2012a; Casper et al., 2013), and so the levels we proposed with Chinook salmon may be broadly applicable to other species, although more studies are needed because there is so much variability in fish ear morphology.

25.7.3 The Significance of Applied Studies

Although the work described starts to contribute to understanding of the effects of man-made sounds on fishes, this is an area ripe for further study that has worldwide implications as shipping and the exploration for traditional and alternative energy sources increase (see Popper & Hawkins, 2012). Indeed, the interest in this area is shown by the very high participation in international meetings that my friend and colleague Tony Hawkins and I have been organizing over the past several years (e.g., Popper & Hawkins, 2012).

From the perspective of someone who has spent more than 45 years studying fish hearing and the evolution of hearing in vertebrates, moving into the applied arena is a big change. However, what I have learned (and what Dick Fay and I expressed in our 2012 paper) is that there are unique opportunities afforded by delving into applied questions. Not only does one get access to nontraditional funding sources, but there are also creative ways to use the applied research to continue to explore important basic science questions (such as hearing capabilities and sound source localization). Moreover, one gets a chance to use what one has learned over decades to help solve problems that are important for the environment and for humans.

25.8 Final Thoughts

In fact, the past years have seen increases in our understanding of fish hearing in the evolution of vertebrate hearing. Still, there are a myriad of open questions (also see Chapter 14 by Hawkins), starting with my favorite, sound source localization, but

including numerous comparative questions, some of which were raised in this chapter. It will be exciting to see what the next generation of investigators finds. But, at the same time, I anticipate that many of these discoveries will be made in the context of solving problems that have a "translational" or applied aspect.

Acknowledgments I have been truly fortunate over the last 40+ years to have a large number of great collaborators, including a wonderful group of students and postdocs. I would very much have liked to mention the work of every one of these people, but because Dick and I have limited the lengths of the chapters in this volume, I had to focus on just a few of the many questions that my lab has dealt with over the years. I do want all those students and collaborators not mentioned here to know, however, that I value and appreciate working with every one of them and that perhaps, in the future, I can discuss their work. I also thank the many funding agencies who have, for 43 years without interruption, supported the work in my lab. I also want to thank Allison Coffin, Sheryl Coombs, Brandon Casper, and Christopher Platt for reviewing the manuscript, and my oldest friend, Steve Weinberg (from Mr. Vinci's sixth grade class), for asking penetrating questions that helped me improve the manuscript substantially. Finally, I am most grateful to my wife Helen for editing this manuscript—she has not only totally supported me in my work over the years, but she is also the very best editor, and I am grateful for her sharp eye and pen.

References

Breder, C. M. Jr. (1943). Problems in the behavior and evolution of a species of blind cave fish. *Transactions of the New York Academy of Sciences*, 5, 168–176.

Buran, B. N., Deng, X., & Popper, A. N. (2005). Structural variation in the inner ears of four deep-sea elopomorph fishes. *Journal of Morphology*, 265, 215–225.

Cahn, P. H. (1958). Comparative optic development in *Astyanax mexicanus* and in two of its blind cave derivatives. *Bulletin of the American Museum of Natural History*, 115, 75–112.

Cahn, P. H., Ed. (1967). *Lateral line detectors*. Bloomington: Indiana University Press.

Casper, B. M., Smith, M. E., Halvorsen, M. B., Sun, H., Carlson, T. J., & Popper, A. N. (2013). Effects of exposure to pile driving sounds on fish inner ear tissues. *Comparative Biochemistry and Physiology A*, 166, 352–360.

Coombs, S., & Popper, A. N. (1979). Hearing differences among Hawaiian squirrelfishes (Family Holocentridae) related to differences in the peripheral auditory system. *Journal of Comparative Physiology*, 132, 203–207.

Corwin, J. T. (1981a). Audition in elasmobranchs. In W. N. Tavolga, A. N. Popper, & R. R. Fay (Eds.), *Hearing and sound communication in fishes*, (pp. 81–105). New York: Springer-Verlag.

Corwin, J. T. (1981b). Postembryonic production and aging in inner ear hair cells in sharks. *Journal of Comparative Neurology*, 201, 541–553.

Dale, T. (1976). The labyrinthine mechanoreceptor organs of the cod *Gadus morhua* L. (Teleostei, Gadidae). *Norwegian Journal of Zoology*, 24, 85–128.

Deng, X., Wagner, H.-J., & Popper, A. N. (2011). The inner ear and its coupling to the swim bladder in the deep-sea fish *Antimora rostrata* (Teleostei: Moridae). *Deep Sea Research, Part I*, 58, 27–37.

Deng, X., Wagner, H.-H., & Popper, A. N. (2013). Interspecific variations of inner ear structure in the seep-sea fish family Melamphaidae. *Anatomical Record*, 296, 1064–1082. doi: 10.1002/ar.22703.

Denton, E. J., & Gray, J. A. (1979). The analysis of sound by the sprat ear. *Nature*, 282, 406–407.

Dijkgraaf, S. (1932). Untersuchunger über die Funktion der Seitenorgane an Fischen. *Zeitschrift für vergleichende Physiologie*, 20, 162–214.

Dunning, D. J., Ross, Q. E., Geoghegan, P., Reichle, J. J., Menezes, J. K., & Watson, J. K. (1992). Alewives avoid high-frequency sound. *North American Journal of Fisheries Management*, 12(3), 407–416.

Fay, R. R., & Popper, A. N. (2000). Evolution of hearing in vertebrates: The inner ears and processing. *Hearing Research*, 149, 1–10.

Fay, R. R. & Popper, A. N. (2012). Fish hearing: New perspectives from two "senior" bioacousticians. *Brain, Behaviour and Evolution*, 792, 215–217.

Halvorsen, M. B., Casper, B. C., Matthews, F., Carlson, T. J., & Popper, A. N. (2012a). Effects of exposure to pile driving sounds on the lake sturgeon, Nile tilapia, and hogchoker. *Proceedings of the Royal Society of London B: Biological Sciences*, 279, 4705–4714.

Halvorsen, M. B., Casper, B. M., Woodley, C. M., Carlson, T. J., & Popper, A. N. (2012b). Threshold for onset of injury in Chinook salmon from exposure to impulsive pile driving sounds. *PLoS ONE*, 7(6) e38968.

Higgs, D. M., Souza, M. J., Wilkins, H. R., Presson, J. C., & Popper, A. N. (2002). Age- and size-related changes in the inner ear and hearing ability of the adult zebrafish (*Danio rerio*). *Journal of the Association for Research in Otolaryngology*, 3, 174–184.

Higgs, D. M., Plachta, D. T. T., Rollo, A. K., Singheiser, M., Hastings, M. C., & Popper, A. N. (2004). Development of ultrasound detection in of American shad (*Alosa sapidissima*). *Journal of Experimental Biology*, 207, 155–163.

Jacobs, D. W., & Tavolga, W. N. (1967). Acoustic intensity limens in the goldfish. *Animal Behaviour*, 15, 324–335.

Ladich, F., & Fay, R. R. (2013). Auditory evoked potential audiometry in fish. *Reviews in Fish Biology and Fisheries* 23, 317–364. doi:10.1007/s11160-012-9297-z

Lombarte, A., & Popper, A. N. (1994). Quantitative analyses of postembryonic hair cell addition in the otolithic endorgans of the inner ear of the European hake, *Merluccius merluccius* (Gadiformes, Teleostei). *Journal of Comparative Neurol*ogy, 345, 419–428.

Lombarte, A. Yan, H. Y., Popper, A. N., Chang, J. C., & Platt, C. (1993). Damage and regeneration of hair cell ciliary bundles in a fish ear following treatment with gentamicin. *Hearing Research*, 66, 166–174.

Mann, D. A., Lu, Z., & Popper, A. N. (1997). A clupeid fish can detect ultrasound. *Nature*, 389, 341.

Mann, D. A., Wilson, C. D., Song, J., & Popper, A. N. (2009). Hearing sensitivity of the Walleye Pollock, *Theragra chalcogramma. Transactions of the American Fisheries Society*, 138, 1000–1008.

Mathiesen, C., & Popper, A. N. (1987). The ultrastructure and innervation of the ear of the gar, *Lepisosteus osseus. Journal of Morphology*, 194, 129–142.

McCauley, R. D., Fewtrell, J., & Popper, A. N. (2003). High intensity anthropogenic sound damages fish ears. *Journal of the Acoustical Society of America*, 113, 638–642.

McCormick, C. A., & Popper, A. N. (1984). Auditory sensitivity and psychophysical tuning curves in the elephant nose fish, *Gnathonemus petersii. Journal of Comparative Physiology*, 155, 753–761.

McPhee, J. (2003). *The founding fish*. New York: Farrar, Straus and Giroux.

O'Connell, C. P. (1955). The gas bladder and its relation to the inner ear in *Sardinops caerulea* and *Engraulis mordax. Fishery Bulletin*, 56, 505–533.

Parker, G. H. (1902). Hearing and allied senses in fishes. *Bulletin of the U.S. Fisheries Commission*, 22, 45–64.

Parvulescu, A. (1964). Problems of propagation and processing. In W. N. Tavolga (Ed.), *Marine bio-acoustics* (pp. 87–100). Oxford: Pergamon Press.

Plachta, D. T. T., & Popper, A. N. (2003). Evasive responses of American shad (*Alosa sapidissima*) to ultrasonic stimuli. *Acoustical Research Letters Online*, 4, 25–30.

Plachta, D. T. T., Song, J., Halvorsen, M. B., & Popper, A. N. (2004). Neuronal encoding of ultrasonic sound by a fish. *Journal of Neurophysiology*, 91, 2590–2597.

Platt, C. (1977). Hair cell distribution and orientation in goldfish otolith organs. *Journal of Comparative Neurology*, 172, 283–297.

Platt, C. (1983). The peripheral vestibular system in fishes. In R. G. Northcutt & R. E. Davis (Eds.), *Fish neurobiology* (pp. 89–124). Ann Arbor: University of Michigan Press.

Platt, C. (1993). Zebrafish inner ear sensory surfaces are similar to those in goldfish. *Hearing Research*, 63, 133–140.

Platt, C., & Popper, A. N. (1984). Variation in lengths of ciliary bundles on hair cells along the macula of the sacculus in two species of teleost fishes. *Scanning Electron Microscopy*, 1984, 1915–1924.

Platt, C., Jørgensen, J. M., & Popper, A. N. (2004). The inner ear of the lungfish *Protopterus*. *Journal of Comparative Neurology*, 471, 277–278.

Pliny (The Elder) (1890). *The natural history of Pliny*, Vol. II. Translated by J. Bostock and H. T. Riley, London: George Bell & Sons. Available at http://books.google.com/books?id=BUEMAAAAIAAJ&printsec=frontcover&source=gbs_ge_summary_r&cad=0#v=onepage&q=clapping&f=false

Popper, A. N. (1970). Auditory capacities of the Mexican blind cave fish *Astyanax jordani* and its eyed ancestor *Astyanax mexicanus*. *Animal Behaviour*, 18, 52–562.

Popper, A. N. (1971). The morphology of the Weberian ossicles in two species of *Astyanax* (Ostariophysi: Characidae). *Journal of Morphology*, 133, 179–188.

Popper, A. N. (1976). Ultrastructure of the auditory regions in the inner ear of the lake whitefish. *Science*, 192, 1020–1023.

Popper, A. N. (1977). A scanning electron microscopic study of the sacculus and lagena in the ears of fifteen species of teleost fishes. *Journal of Morphology*, 153, 397–418.

Popper, A. N. (1978). Scanning electron microscopic study of the otolithic organs in the bichir (*Polypterus bichir*) and shovel-nose sturgeon (*Scaphirhynchus platorynchus*). *Journal of Comparative Neurology*, 18, 117–128.

Popper, A. N. (1980). Scanning electron microscopic studies of the sacculus and lagena in several deep-sea fishes. *American Journal of Anatomy*, 157, 115–136.

Popper, A. N. (1981). Comparative scanning electron microscopic investigations of the sensory epithelia in the teleost sacculus and lagena. *Journal of Comparative Neurology*, 200, 357–374.

Popper, A. N., & Coombs, S. (1982). The morphology and evolution of the ear in Actinopterygian fishes. *American Zoologist*, 22, 311–328.

Popper, A. N., & Fay, R. R. (1993). Sound detection and processing by fish: Critical review and major research questions. *Brain Behaviour and Evolution*, 41, 14–38.

Popper, A. N., & Fay, R. R. (2011). Rethinking sound detection by fishes. *Hearing Research*, 273, 25–36.

Popper, A. N., & Hastings, M. C. (2009). Effects of anthropogenic sources of sound on fishes. *Journal of Fish Biology*, 75, 455–498.

Popper, A. N., & Hawkins, A. D., Eds. (2012). *The effects of noise on aquatic life*. New York: Springer Science+Business Media.

Popper, A. N., & Hoxter, B. (1984). Growth of a fish ear: 1. Quantitative analysis of sensory hair cell and ganglion cell proliferation. *Hearing Research*, 15, 133–142.

Popper, A. N., & Northcutt, R. G. (1983). Structure and innervation of the inner ear of the bowfin, *Amia calva*. *Journal of Comparative Neurology*, 21, 279–286.

Popper, A. N., & Platt, C. (1979). The herring ear has a unique receptor pattern. *Nature*, 280, 832–833.

Popper, A. N., & Platt, C. (1983). Sensory surface of the saccule and lagena in the ears of ostariophysan fishes. *Journal of Morphology*, 176, 121–129.

Popper, A. N., Fay, R. R., Platt, C., & Sand, O. (2003). Sound detection mechanisms and capabilities of teleost fishes. In S. P. Collin & N. J. Marshall (Eds.), *Sensory processing in aquatic environments* (pp. 3–38). New York: Springer Science+Business Media.

Popper, A. N., Smith, M. E., Cott, P. A., Hanna, B. W., MacGillivray, A. O., Austin, M. E., & Mann, D. A. (2005). Effects of exposure to seismic airgun use on hearing of three fish species. *Journal of the Acoustical Society of America*, 117, 3958–3971.

Ramcharitar, J., Gannon, D. P., & Popper, A. N. (2006). Bioacoustics of the family Sciaenidae (croakers and drumfishes). *Transactions of the American Fisheries Society*, 135, 1409–1431.

Retzius, G. (1881). *Das Gehörorgan der Wirbelthiere*. Vol. I. Stockholm: Samson and Wallin.

Roeder, K. D., & Treat, A. E. (1961). The detection and evasion of bats by moths. *Readings in the Psychology of Perception*, 49, 35.

Rogers, P. H., & Cox, M. (1988). Underwater sound as a biological stimulus. In J. Atema, R. R. Fay, A. N. Popper & W. N. Tavolga (Eds.), *Sensory biology of aquatic animals* (pp. 131–149). New York: Springer-Verlag.

Rogers, P. H., & Zeddies, D. G. (2008). Multipole mechanisms for directional hearing in fish. In J. F. Webb, A. N. Popper, & R. R. Fay (Eds.), *Fish bioacoustics* (pp. 233–252). New York: Springer Science+Business Media.

Rogers, P. H., Popper, A. N., Cox, M., & Saidel, W. M. (1988). Processing of acoustic signals in the auditory system of bony fish. *Journal of the Acoustical Society of America*, 83, 338–349.

Smith, M. E., Coffin, A. B., Miller, D. L., & Popper, A. N. (2006). Anatomical and functional recovery of the goldfish (*Carassius auratus*) ear following noise exposure. *Journal of Experimental Biology*, 209, 4193–4202.

Song, J., Mann, D. A., Cott, P. A., Hanna, B. W., & Popper, A. N. (2008). The inner ears of northern Canadian freshwater fishes following exposure to seismic air gun sounds. *Journal of the Acoustical Society of America*, 124, 1360–1366.

Tavolga, W. N., Ed. (1964). *Marine bio-acoustics*. Oxford: Pergamon Press.

Tavolga, W. N., Ed. (1967). *Marine bio-acoustics*, II. Oxford: Pergamon Press.

Tavolga, W. N., & Wodinsky, J. (1963). Auditory capacities in fishes. Pure tone thresholds in nine species of marine teleosts. *Bulletin of the American Museum of Natural History*, 126, 177–240.

van Bergeijk, W. A. (1964). Directional and nondirectional hearing in fish. In W. N. Tavolga (Ed.). *Marine bio-acoustics* (pp. 281–289). Oxford: Pergamon Press.

von Békésy, G. (1967). Some similarities in sensory perception of fish and man. In P. H. Cahan (Ed.). *Lateral line detectors* (pp. 417–435). Bloomington: Indiana University Press.

von Frisch, K. (1923). Ein Zwergwels der kommt, wenn man ihm pfeift. *Biology Zentralblatt Leipzig*, 43, 439–446.

Weber, E. H. (1820). *De Aure et Auditu Hominis et Animalium. Pars I. De Aure Animalium Aquatilium.* Leipzig: Gerhard Fleischer.

Webster, D. B. (1962). A function of the enlarged middle-ear cavities of the kangaroo rat, *Dipodomys. Physiological Zoology*, *35*(3), 248–255.

Webster, D. B., Fay, R. R., & Popper, A. N., Eds. (1992). *Evolutionary biology of hearing*, New York: Springer-Verlag.

Wersäll, J., Flock, Å., & Lundquist, P. G. (1965). Structural basis for directional sensitivity in cochlear and vestibular sensory receptors. *Cold Spring Harbor Symposium on Quantitative. Biology*, 30, 115–132.

Wilson, M., Montie, E. W., Mann, K. A., & Mann, D. A. (2009). Ultrasound detection in the Gulf menhaden requires gas-filled bullae and an intact lateral line. *Journal of Experimental Biology*, 212(21), 3422–3427.

Yan, H. Y., Saidel, W. M., Chang, J., Presson, J. C., & Popper, A. N. (1991). Sensory hair cells of the fish ear: evidence of multiple types based on ototoxicity sensitivity. *Proceedings of the Royal Society of London B: Biological Sciences*, 245, 133–138.