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Form and Function in the Unique Inner Ear of a Teleost: The Silver Perch (*Bairdiella chrysoura*)

JOHN UMAR RAMCHARITAR,[1]* XIAOHONG DENG,[1] DARLENE KETTEN,[2]
AND ARTHUR N. POPPER[1]
[1]Department of Biology, University of Maryland, College Park, Maryland 20742
[2]Woods Hole Oceanographic Institution, Woods Hole, Massachusetts 02543

ABSTRACT

Members of the teleost family Sciaenidae show significant variation in inner ear and swim bladder morphology as well as in the relationship between the swim bladder and the inner ear. In the silver perch (*Bairdiella chrysoura*), a *Stellifer*-group sciaenid, both the saccular and utricular otoliths are enlarged relative to those in other teleosts. Additionally, its swim bladder is two-chambered, and the anterior chamber surrounds the otic capsule and terminates lateral to the saccules. Structure and function of the auditory system of the silver perch were explored by using gross dissections, scanning electron microscopy, CT scan reconstruction, and auditory brainstem response approach. Several morphological specializations of the auditory system of the silver perch were found, including expansion of the utricular and lagenar otoliths, close proximity between the saccules and the utricles, deeply grooved sulci on the saccular otoliths, two-planar saccular sensory epithelia, and a unique orientation pattern of sensory hair cell ciliary bundles on the saccular sensory epithelium. It was determined that the silver perch can detect up to 4 kHz, with lowest auditory thresholds between 600 Hz and 1 kHz. Audition in the silver perch is comparable to that in the goldfish (*Carassius auratus*), a hearing "specialist." The morphological specializations of the inner ear and swim bladder of the silver perch may be linked to its enhanced hearing capabilities. The findings of this study support the proposal that sciaenids are excellent model species for investigating structure–function relations in the teleost auditory system. J. Comp. Neurol. 475:531–539, 2004. © 2004 Wiley-Liss, Inc.

Indexing terms: audition; ciliary bundle; inner ear; otolith; sciaenid

Animal models have played a critical role in our understanding of the relationship between structure and function in the auditory system of humans (Friedman et al., 1999; Ernest et al., 2000; Bermingham-McDonogh and Rubel, 2003). Although tremendous diversity exists in inner ear anatomy among vertebrates (see, e.g., Retzius, 1881; Wever, 1974), there is at the same time significant similarity in its form and function across taxa (Fay and Popper, 2000). It has been demonstrated, for example, that teleost fishes possess specializations of their hearing system similar to those found in amniotes, e.g., sensory hair cell heterogeneity (Sugihara and Furukawa, 1989; Lanford et al., 2000).

Teleost fishes show substantial interspecific variation in their inner ear structure and function, including size and shape of the otolithic end organs, orientation patterns of the sensory hair cells, peripheral auditory structures, distribution of different sensory hair cell types, frequency detection range, and hearing sensitivity (Fay, 1988; Popper and Fay, 1993; Popper and Platt, 1993; Popper and Lu, 2000). Considerable variability in structure and function of the auditory system is found not only in comparing broad taxonomically distinct fishes but also even within several families of teleosts, including Sciaenidae (drumfishes) and Holocentridae (squirrelfishes; Nelson, 1955;

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*Correspondence to: John Umar Ramcharitar, Department of Psychological and Brain Science, Johns Hopkins University, 3400 N. Charles St., Baltimore, MD 21218. Email: jramch@jhu.edu

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Popper, 1977; Chao, 1978; Davorec, 1983; Ramcharitar et al., 2002).

Among sciaenid fishes, there is significant variation in inner ear and swim bladder morphology as well as in the relationship between the swim bladder and the inner ear (Nelson, 1955; Chao, 1978; Coombs and Popper, 1979; Davorec, 1983; Ramcharitar et al., 2002). For example, whereas the saccular otoliths in all sciaenids are enlarged (compared with those in the vast majority of nonsciaenid species), their shapes and sizes are very different from one species to another (Chao, 1978; Davorec, 1983; Ramcharitar, 2003). Additionally, swim bladder anatomy and proximity to the inner ear vary between species (Chao, 1978; Davorec, 1983; Ramcharitar, 2003).

Among the 11 supragenera of western Atlantic sciaenids defined by Chao (1978), the *Stellifier* group, of which the silver perch is a member, shows the most morphological specialization in the hearing system. First, their utricular otoliths, like their saccular otoliths, are enlarged. The *Stellifer* group members also possess segmented swim bladders, with yoke-shaped anterior segments and a "carrot-shaped" posterior segment (Chao, 1978). In the silver perch, the anterior swim bladder segment terminates just lateral to the saccules (Ramcharitar, 2002).

The relationship between the swim bladder and the otic capsule, as well as otolith morphology, is believed to be an important contributor to fish audition. When the swim bladder is in close proximity to the inner ear, it behaves as a peripheral auditory structure by expanding the frequency range of detection as well as by improving auditory sensitivity (Fay and Popper, 1974; Denton and Blaxter, 1976; Blaxter and Tytler, 1978). Swim bladders and other gas chambers are pressure detectors that reradiate acoustic energy as particle displacements, which can potentially stimulate the inner ear if the two structures are close together (van Bergeijk, 1967; Kalmijn, 1988; Rogers and Cox, 1988). In contrast, otoliths may affect audition, in that their size and shape are thought to influence the ability to respond to acoustic stimuli; e.g., greater otolithic mass is predicted to confer higher sensitivity to low-frequency sounds (DeVries, 1950; Grant and Best, 1987; Lychakov and Rebane, 1993).

Ramcharitar et al. (2002) demonstrated that sciaenid species with swim bladders closely associated with the otic capsule (spotted seatrout, *Cynoscion nebulosus*; and Atlantic croaker, *Micropogonias undulatus*) have larger numbers of stereocilia per ciliary bundle on the sensory hair cells of the saccule, as well as greater rostral expansion of the saccular sensory epithelium, compared with species in which the swim bladder is distant from the ear (spot, *Leiostomus xanthurus*; and kingfish, *Menticirrhus americanus*). These correlations were also shown to extend to function, insofar as two sciaenids with swim bladders that reach near the otic capsule (weakfish, *Cynoscion regalis*; and Atlantic croaker, *M. undulatus*) have extended frequency ranges of detection compared with a species lacking a close inner ear–swim bladder relationship (spot, *L. xanthurus*; Ramcharitar, 2003).

In this study, we explored structure–function relations in the silver perch, a *Stellifer*-group sciaenid. It was hypothesized that the silver perch will demonstrate better audition than non-*Stellifer*-group sciaenids on account of its inner ear and swim bladder morphological specializations.

MATERIALS AND METHODS

All animals used in this study were supplied by the Public Service Enterprise Group of New Jersey. Fish were kept in captivity in large, cylindrical tanks (890 liters, diameter 122 cm). Water was filtered and aerated, and fish were fed daily until used in the study. Twenty-five individuals were used in this study, which was approved by the University of Maryland Institutional Animal Care and Use Committee. Fish used for morphological analysis were 12–17 cm (TL), whereas those used for investigating hearing ability were 12–15 cm (TL). No significant differences resulting from fish size were found ($P > .05$).

Gross morphology

Five fish were killed with buffered ethyl 3-aminobenzoate methanesulfonate (MS-222; Sigma), and ventral dissections were performed to expose the swim bladder and otic capsule. Digital images of the swim bladder as well as the relationship between the swim bladder and the otic capsule were then acquired. Otoliths were extracted and examined with a dissecting microscope and subsequently visualized by scanning electron microscopy (SEM). Three-dimensional reconstruction of otoliths from CT scan analysis was performed in the laboratory of Dr. D. Ketten in Woods Hole, Massachusetts.

Scanning electron microscopy

Before dissection, fish were killed with MS-222 (Sigma) and then fixed in 5% paraformaldehyde (in 0.1 M phosphate buffer, pH 7.4) after the heads were opened by dissection to expose the ears to facilitate penetration of fixative into the ear tissues. After the fixation period, ears were washed with phosphate buffer and stored in 70% ethanol. Ears were postfixed after initial fixation with 2% osmium tetroxide after removal from the head, and sensory epithelia were then removed from the otolithic end organs. Immediately prior to critical-point drying, isolated epithelia were dehydrated through a series of ethanols of increasing concentrations (70%, 85%, 100%). The epithelia were then critical-point dried, with carbon dioxide as the intermediate fluid. The dried specimens were coated with a gold-palladium alloy under high vacuum using a DV503 system and examined on an Amray (model 1820D) scanning electron microscope.

Saccular, lagenar, and utricular hair cell orientations were mapped for each sensory epithelium by using SEM. Ciliary bundle orientation was defined with the axis of polarity pointing to the end of the bundle containing the kinocilium. Low-power images (×25 to ×50) were first taken, and mosaics were then constructed to compose complete pictures of the entire maculae. Bundle orientation was determined for selected regions across each macula by using high-power SEM images (×3,000).

Auditory brainstem response

Prior to experimentation, the position of the brainstem was determined by using gross dissections. Three fish were killed with buffered MS-222 (Sigma), and dorsal dissections were performed to expose the brain and rostral region of the spinal cord. Once the brainstem was mapped, landmarks on the external surface of the fishes were identified to make electrode placement consistent between auditory brainstem response (ABR) experiments.

Tank setup

Fish were suspended in a mesh sling into a 19-liter plastic bucket filled with water. They were positioned about 35 cm above an underwater speaker (UW-30; Underwater Sound Inc., Oklahoma City, OK) and fully submerged at 10 cm below the surface. A reference electrode was placed on the dorsal surface of the fish's head, medial to and just behind the eyes. A recording electrode was placed on the dorsal surface of the fish, just over the brainstem. Both of these electrodes were inserted just beneath the skin to improve the signal-to-noise ratio. The electrodes used were stainless steel (Rochester Electro-Medical Inc., Tampa, FL) insulated with epoxy, except for the very tip (1 mm).

Stimulus generation and signal acquisition

Sounds were played and responses were collected by using a Tucker-Davis Technologies Inc. (TDT, Gainesville, FL) physiological apparatus with SigGen and BioSig software (TDT). Sounds were played from a computer with the TDT software through a power amplifier connected to the UW-30 underwater speaker. Calibration of the acoustic output to the test tank was performed by analyzing the speaker output at the position of the fish at each frequency with an LC-10 hydrophone (calibration sensitivity of –208.6 dB re: 1 V/μPa; ±3 dB, 0.1–180 kHz, omnidirectional) attached to a digital oscilloscope. Additionally, a calibrated hydrophone (InterOcean model 902 Acoustic Listening and Calibration System) was used to double check the speaker output at the test frequencies and sound intensity levels. Pure tones were played at 100-Hz intervals from 100 to 1,500 Hz and then at 500-Hz intervals up to 6 kHz for all fishes. Tones were 10 msec in duration with a 2-msec rise and fall time. Sound intensity levels at each frequency were increased in 5-dB steps until a typical ABR waveform was observed (Fig. 1). In total, 400 responses were averaged for each signal tested.

Controls

Throughout the experimental testing, the ABR system was calibrated by using goldfish, a species that has been studied extensively and for which there are established audiograms (for review see Fay, 1988). The goldfish audiogram is therefore a good standard with which to ensure that our data from the ABR method fit with the general range of hearing sensitivity of established species (Higgs et al., 2002). In each test case, expected audiograms were achieved for this teleost, confirming that our ABR methods were reliable and that our silver perch data could be compared with data from other species in the literature. Experiments were also performed with dead fish or no fish in the setup to establish that the identified ABR responses were not artifacts. These tests also confirmed that our data were reliable.

Statistical analysis

All statistical analyses were performed as analyses of variance (ANOVA), with the Tukey post hoc test when significant differences were found (Zar, 1984). For all tests, $\alpha = .05$ was used as the significance level.

Photomicrograph production

Images were stored and manipulated in the software programs Adobe Photoshop 6.0 and Adobe PageMaker 6.0.

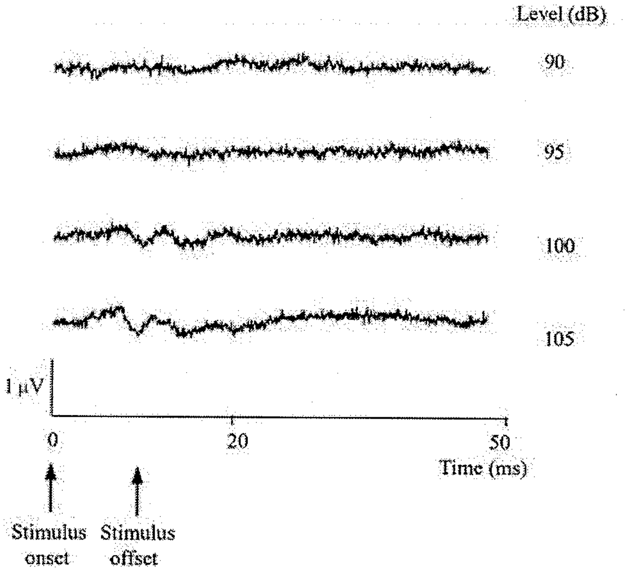

Fig. 1. Typical ABR traces of silver perch for a test frequency of 2.5 kHz. The auditory threshold is here defined as the minimal sound intensity level at which an ABR is just detectable. The response is composed of a series of negative peaks generally within the first 20 msec of stimulus onset. The threshold in this case is judged to be 95 dB. Sound intensity levels are stated in dB (re: 1 μPa).

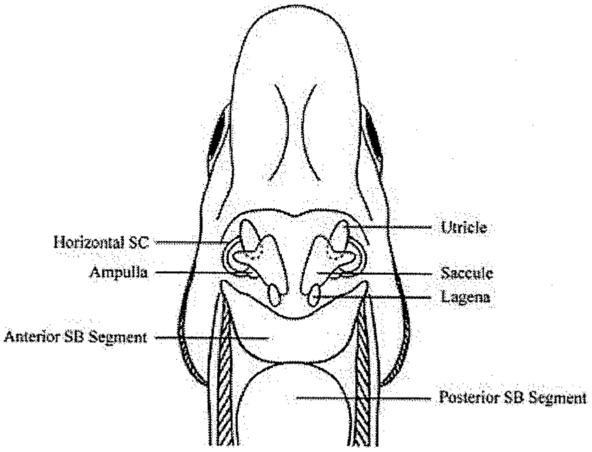

Fig. 2. Diagram of ventral dissection of silver perch showing gross structure of inner ear and swim bladder. Rostral is to the top. SB, swim bladder; SC, semicircular canal.

RESULTS

Swim bladder–inner ear relationship

Figure 2 shows a diagram of a ventral dissection of the silver perch with details of the otolithic end organs and swim bladder. The anterior segment of the swim bladder curves around the caudal region of the otic capsule and terminates lateral to the saccules. The closest approach of the swim bladder to the inner ear is at the posterior edge of the otic capsule near the lagenae. Also, the terminal ends of the anterior swim bladder segment lie just be-

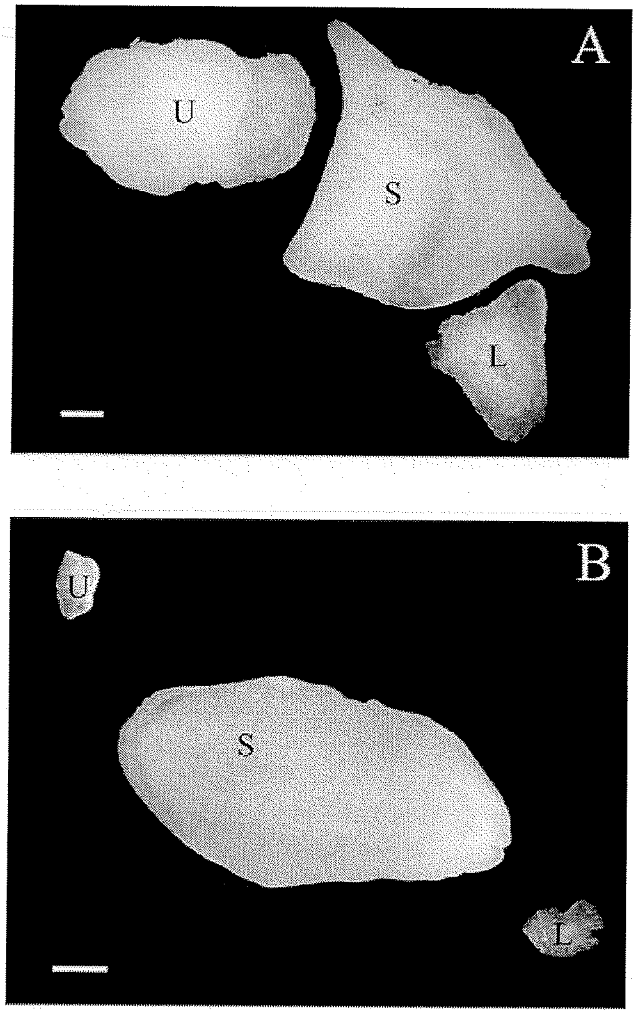

Fig. 3. Relative shapes and sizes of the otoliths of silver perch (*Stellifer* species) and weakfish (non-*Stellifer* species). **A:** Inner surfaces of the otoliths of silver perch (17 cm TL). **B:** Inner surfaces of the otoliths of weakfish (16.5 cm TL). Rostral is to the left, and dorsal is to the top. U, utricular otolith; S, saccular otolith; L, lagenar otolith. Scale bars = 1 mm.

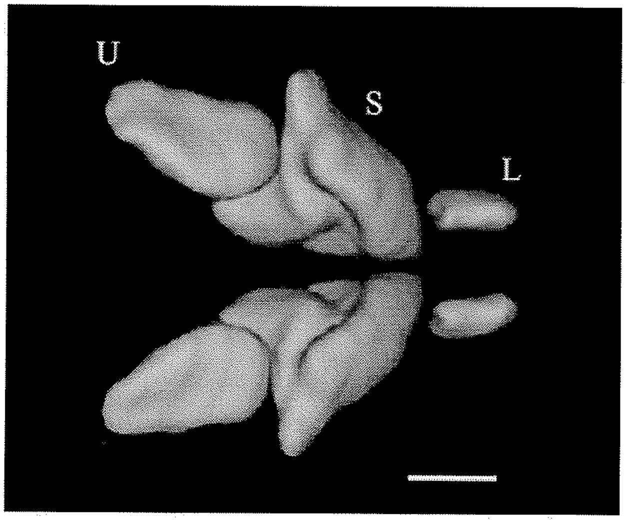

Fig. 4. Three-dimensional CT reconstruction of silver perch otoliths in dorsal view showing relative shapes, sizes, and positions in space. Rostral is to the left. U, utricle; S, saccule; L, lagena; R, rostral. Artifacts were removed in Adobe Photoshop. The specimen used was 18 cm long (TL). Scale bar = 5 mm.

neath the skin and close to a region where several lateral line canals were found.

Otolith morphology

The otoliths of the silver perch are enlarged relative to those of other non-*Stellifer*-group sciaenids investigated (Fig. 3A). The saccular and lagenar otoliths of the silver perch are thick and triangular, whereas those associated with the utricles are thick and oval. The otoliths of a non-*Stellifer*-group sciaenid, the weakfish, are presented in Figure 3B for comparison with those of the silver perch. Both fish are of the same relative size (silver perch TL = 17.0 cm, weakfish TL = 16.5 cm). In weakfish, only the sagittae are enlarged, and they are oval, elongated, and moderately thick. The utricular and lagenar otoliths of weakfish are significantly smaller than those of the silver perch. An additional otolithic difference was observed in the sulci of the lagenar otoliths. In the weakfish and other non-*Stellifer* sciaenids, only the ventral portion of the lagenar sensory epithelium contacts the otolith, whereas in the silver perch the entire sensory epithelium interacts with the otolith.

Three-dimensional arrangement of otoliths

Figure 4 is a three-dimensional reconstruction (by using CT analysis) of the otoliths of silver perch (TL = 18 cm) showing their relative sizes and positions in space (dorsal view). The lower caudal region of the utricular otolith is in close proximity to the upper rostral portion of the saccular otolith. To a lesser extent, the lagenar otoliths and sagittae are also in close proximity to each other.

Sensory epithelia

The saccular sensory epithelium of silver perch is a three-dimensional structure (Fig. 5). Figure 5A shows the relationship between this epithelium and the saccular otolith. The sulcus of the saccular otolith in which the sensory epithelium rests is deeply grooved in this species. The saccular sensory epithelium is positioned in two planes, one vertical and the other at an angle of about 120° from the vertical plane (Fig. 5B).

In the vertical portion of the saccular epithelium, the ciliary bundles form two distinct populations along the midline of the epithelium, and they are oriented away from the dividing line and at about right angles to it. The other segment of the epithelium also has two distinct populations about its long axis, and they are generally oriented toward the dividing line, except at the rostral tip (Fig. 6A′).

The utricular sensory epithelium, like that associated with the saccule, is a three-dimensional structure. It is "bowl shaped," with an elongate lacinia present (Fig. 6B). The anteriolateral band of ciliary bundles on this epithelium has cells directed posteriomedially, whereas those

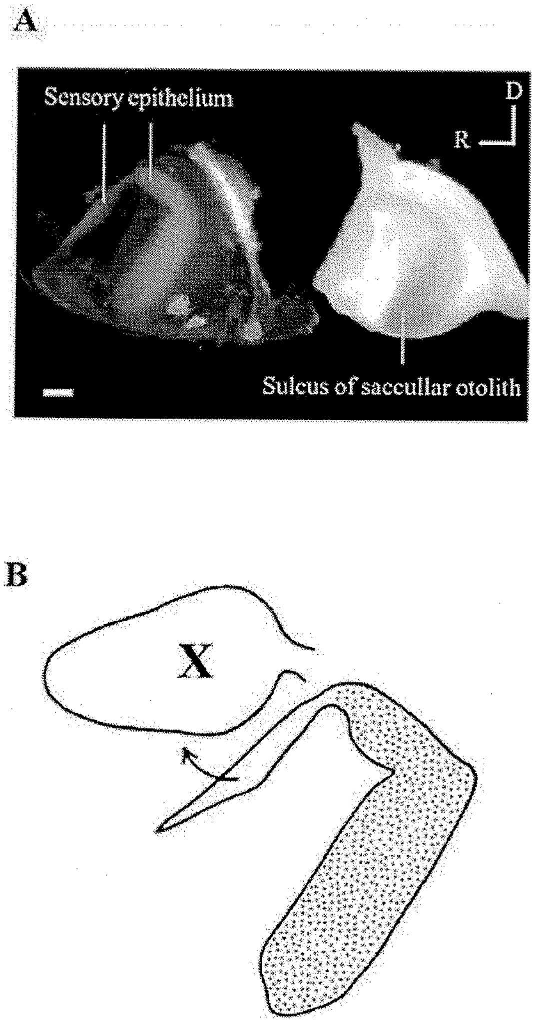

Fig. 5. Saccular sensory epithelium of silver perch. **A:** Relationship between the saccular epithelium and the otolith. The sulcus of the otolith is deeply grooved and extends dorsally over the otolith, forming two planes at about 120° to each other. The epithelium shown was lifted from this sulcus (sensory hair cell ciliary bundles on the epithelium face toward the sulcus). **B:** Diagram showing the shape of the saccular epithelium. Stippled region lies in the vertical plane. X, flattened view of segment of epithelium that curves over the otolith; D, dorsal; R, rostral. Scale bar = 1 mm.

that populate the central region are oriented in the anteriolateral direction (Fig. 6B).

The lagenar sensory epithelium is positioned vertically in space, and it possesses ciliary bundles that show a graded orientation pattern from vertical in the upper segment to horizontal in the lower portion of the epithelium. The ciliary bundles on this epithelium are organized in two opposing populations about the longitudinal axis (Fig. 6C).

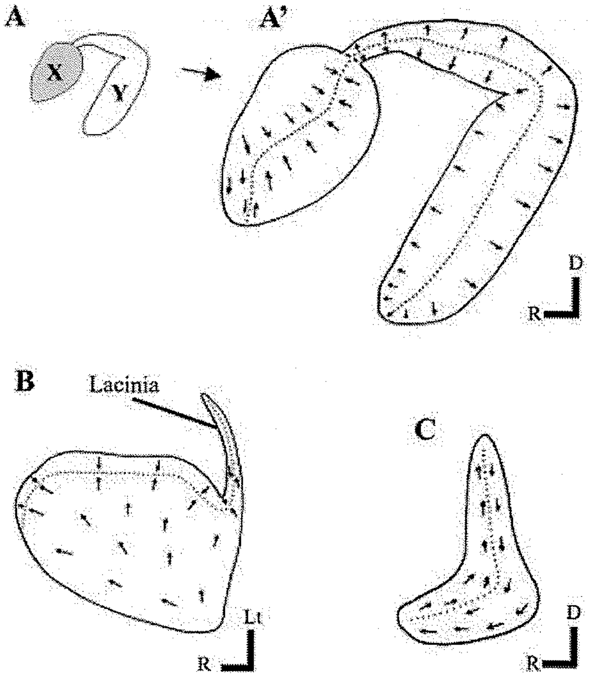

Fig. 6. Schematics of saccular, lagenar, and utricular sensory epithelia showing ciliary bundle orientation patterns. The arrows indicate the direction to which bundles are maximally sensitive to acoustic particle motion. **A:** Two major regions of the saccular epithelium (flattened): Y lies in the vertical plane, and X lies in a plane at 120° to the vertical plane (see Fig. 5). **A′:** Flattened saccular epithelium. **B:** Utricular epithelium. Note the presence of a lacinia that is elongated and oriented laterally. **C:** Lagenar epithelium. R, rostral; D, dorsal; Lt, lateral.

TABLE 1. Auditory Thresholds of Silver Perch[1]

F (Hz)	Mean threshold (dB)	SD
100	86.2	4.63
200	87.9	5.20
300	83.5	4.22
400	83.8	4.13
500	86.2	2.57
600	74.6	5.01
700	82.3	4.09
800	77.8	4.67
900	81.3	5.63
1,000	81.3	4.79
1,100	86.7	2.89
1,200	87.3	3.21
1,300	88.7	5.77
1,500	88.9	6.07
2,000	87.9	6.24
3,000	93.9	6.16
4,000	102.3	2.47

[1] Thresholds are given in dB (re: 1 μPa). F, frequency; SD, standard deviation.

Auditory thresholds

Table 1 summarizes the auditory threshold data for silver perch. This species detected frequencies from 100 Hz to 4 kHz, with best hearing sensitivity between 600 Hz and 1 kHz. There were no significant differences in auditory thresholds for test signals from 100 to 500 Hz ($P >$ .05), and these thresholds were higher than that at 600 Hz ($P <$ .05). Hearing thresholds between 600 Hz and 1 kHz ($P >$ .05) and those between 1.1 and 3 kHz were also statistically alike ($P >$ .05).

Figure 7 shows the audiograms for both silver perch and goldfish. The frequency detection ranges of both species are alike and so are the auditory thresholds except at 400, 500, and 4,000 Hz. At these three test frequencies, gold-

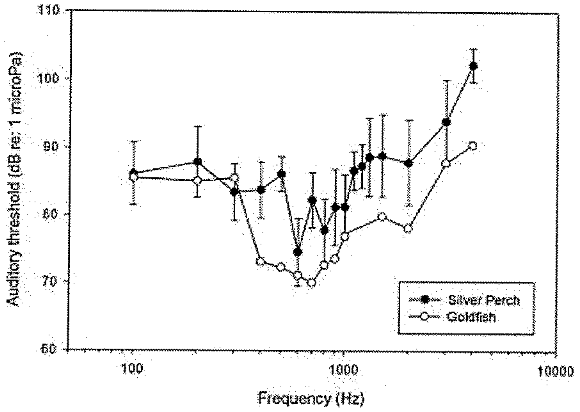

Fig. 7. Audiogram of silver perch. Mean auditory thresholds for goldfish are included for comparison. Silver perch responded to frequencies up to 4 kHz, with best detection between 600 and 1 kHz. Goldfish showed better hearing sensitivity between 400 and 500 Hz, as well as at 4 kHz.

fish showed better sensitivity than silver perch ($P < .05$). The auditory threshold data presented for goldfish were derived from ABR studies in a setup identical to that used for investigating the silver perch.

DISCUSSION

It was shown that the inner ear of the silver perch has a number of specialized structural and functional properties. These specializations of the inner ear and swim bladder of the silver perch have the potential of revealing how specific anatomical characteristics support hearing. First, the silver perch has a unique spatial relationship between the otolithic end organs never before described for a fish or any other vertebrate, i.e., the close proximity of the enlarged saccules and utricles. Additionally unique to silver perch is the shape of the saccular sensory epithelium and its sensory hair cell ciliary bundle orientation pattern. Compared with other non-*Stellifer*-group sciaenids for which data are available, the silver perch has the broadest hearing bandwidth, and this correlates with its unique swim bladder–inner ear relationship as well as with its inner ear anatomy. It is, however, probable that other *Stellifer*-group sciaenids possess auditory specializations identical to those of the silver perch.

It is highly plausible that the unique swim bladder–inner ear configuration of silver perch is responsible for the broad frequency range of detection. Swim bladders have been modeled after highly resonant, underwater, pulsating bubbles (Harris, 1964; van Bergeijk, 1967). As such, the swim bladder may be transforming sound pressure into particle displacements, which can then potentially stimulate the inner ear if it is in close proximity to the otolithic end organs (van Bergeijk, 1967; McCartney and Stubbs, 1971). This is supported in the literature by several studies taking diverse approaches, including swim bladder ablation, artificial gas bladders, behavior, and physiology (see, e.g., Sand and Hawkins, 1973; Chapman and Sand, 1974; Popper, 1974).

Among the sciaenid species previously studied, a correlation between swim bladder–inner ear relationship and frequency range of detection has been found (Ramcharitar, 2003). Weakfish, a species in which the anterior end of the swim bladder terminates within a few millimeters of the otic capsule, is able to detect frequencies up to 2 kHz, whereas spot detects up to 700 Hz and has a swim bladder that is distant from the ear. The Atlantic croaker, a species with a swim bladder–ear relationship that is somewhat intermediate between the relationships in weakfish and spot, is able to detect up to 1 kHz. Hence there is a gradient in hearing ability related to the swim bladder–inner ear configuration between these fishes.

The detection range of silver perch is at least twice that of weakfish, a non-*Stellifer*-group sciaenid that has been designated a "hearing specialist" (Ramcharitar, 2003). In the silver perch, as in weakfish, the anterior swim bladder segment reaches within millimeters of the posterior edge of the otic capusule. However, in the silver perch, as described earlier, the swim bladder also encircles the otic capsule, terminating just lateral to the saccules. This configuration could potentially facilitate complex auditory input from the swim bladder to the otolithic end organs, thereby conferring enhanced hearing on this species. Swim bladder morphology and its relationship with the inner ear may therefore account for the disparity in audition between the silver perch and the weakfish.

It has been proposed that swim bladder anatomy affects the resonant properties (Schellart and Popper, 1992). For example, in elongated gas chambers, the particle motions parallel with the long axis are large, but they attenuate steeply with distance compared with spherical bubbles. Further studies on the resonant properties of sciaenid swim bladders (e.g., using laser vibrometry) will help to elucidate the possible contributions of these organs to audition. Specifically, such studies may help to answer questions on the possible roles of swim bladder projections and segmentation in both audition and vocalization.

In otophysan fishes, the anterior region of the swim bladder is mechanically coupled to the inner ear by a series of bones, the Weberian apparatus (Weber, 1820). The hearing of these fishes is generally up to several kilohertz, with relatively good auditory sensitivity (Fay, 1988). The sound detection capability of silver perch was shown to be similar to that of goldfish, an otophysan species. However, the hearing sensitivity of silver perch between 400 and 700 Hz was relatively poor compared with goldfish, and this may be due to the lack of physical attachment between the swim bladder and the inner ear in the former species. This is supported by the work of Poggendorf (1952), who demonstrated that ablation of the Weberian ossicles in the otophysan catfish *Ictalurus nebulosus* significantly reduces auditory sensitivity across its frequency detection range. Theoretical models and analyses also support these findings (Finneran and Hastings, 2000).

The silver perch has four distinctive specializations of its otoliths: enlargement of its utricular and lagenar otoliths, the triangular shape of its saccular otoliths (sagittae), the deeply grooved sulci of its saccular otoliths, and the close proximity of the saccular and utricular otoliths. In several other teleost species, there are correlations between otolith morphology and audition (see, e.g., Popper and Tavolga, 1981). It is therefore possible that the specializations of otolith structure observed in silver perch may be linked to its enhanced auditory capabilities. These putative otolithic adaptations may also provide mechanisms whereby this species could potentially differentiate between auditory inputs from direct inner ear stimulation via particle displacements in the near field and indirect swim bladder auditory inputs. Such an ability may play a role in sound-source localization (Schuijf and Buwalda, 1975).

Early experimental evidence from lesion and behavioral approaches suggested that the utricle is primarily a vestibular organ (von Frisch, 1938; von Holst, 1950). Among the three otolithic end organs, the utricle is considered to be the most evolutionarily conserved (Flock, 1964; Platt and Popper, 1981). There is evidence, however, that the utricle may also serve an auditory function in some species (Denton et al., 1979; Popper and Tavolga, 1981; Mann et al., 1997, 2001). In the marine catfish (*Arius felis*), the utricle and its associated otolith are both very large compared with those found in other ostariophysans. Additionally, *Arius* shows a surprisingly narrow frequency range of detection but good hearing sensitivity at lower frequencies (Popper and Tavolga, 1981). It was hypothesized that the atypical utricle of this species confers specialized low-frequency audition. Further support for auditory function in the utricle comes from clupeid fishes that have an unusual tripartite utricular macula (Denton and Blaxter, 1976; Blaxter et al., 1981). It has been shown that members of one subfamily of Clupeiformes, the Alosinae, are able to detect ultrasound (Mann et al., 1997, 2001). It is therefore possible that the enlarged utricle of silver perch performs auditory functions. Note that the utricle of this species possesses a thin, elongated lacinia, and this has also been observed for goldfish (*Carassius auratus*), a hearing specialist (Platt, 1977; Platt and Popper, 1981). Lacinias of various shapes and sizes are also present in numerous other teleosts, but their functions are unknown (Platt and Popper, 1981).

Otolith morphology is species specific, with the saccular otoliths showing the most extensive variations (Platt and Popper, 1981). This is exemplified in sciaenids; their sagittae are markedly diverse in form, ranging from oval and thin to shieldlike and very thick (Chao, 1978). In the silver perch, the sagittae are thick and triangular and their deeply grooved sulci curve around the otolith, forming two planes at about 120° to each other. These otolithic specializations probably confer specific types of mechanical properties on the auditory system (DeVries, 1950; Grant and Best, 1987; Lychakov and Rebane, 1993). The nature of the sulcus of the sagittae also has important implications for the placement and directional response patterns of the ciliary bundles in three-dimensional space as well as for the otolith–epithelium interaction. For example, the deeply grooved sulcus of the saccular otolith of the silver perch may imply that the associated epithelium has curved edges, and this may cause the ciliary hair cell bundles to be oriented in a wider range of directions compared with the case when the sulcus is planar and not grooved (as in weakfish). Also, the otolithic membrane may have different physical properties across a grooved sulcus owing to differences in thickness and/or composition, thereby giving rise to differences in ciliary bundle stiffness and hence tuning.

Among teleosts, there are significant interspecific variations in the degree of contact between sensory epithelia and otoliths (Platt and Popper, 1981). Although recent models of otolith function assume that the otolith–epithelium relationship is equivalent across species (Lychakov and Rebane, 1993, 2000), the known differences in this relationship may have important functional consequences. It is theorized that ciliary bundles may be responsive to acoustic particle motion exclusive of the presence of otoliths (Rogers and Cox, 1988), but it is conceivable that direct contact with the otolith produces distinct response properties in such bundles. There might, therefore, be differences in the function of the lagena of the silver perch compared with those of other non-*Stellifer* sciaenids because its sensory epithelium is in complete contact with the otolith, whereas, for the other investigated sciaenids, there is only partial contact. Further neuroanatomical and physiological studies are needed to understand how the otolith–epithelium interaction may affect audition.

The shape and ultrastructure of the saccular sensory epithelium in the silver perch differ from those of the vast majority of teleost species so far investigated, including those within the family Sciaenidae (Popper, 1977, 1978, 1981; Ramcharitar et al., 2002; Ramcharitar, 2003). It is at the same time possible that all *Stellifer* sciaenids share the same auditory specializations. Although substantial interspecific variation in the shape of this epithelium has been reported (Platt and Popper, 1981), it is placed predominantly in the vertical plane. In silver perch, however, this epithelium is oriented primarily in two planes. Therefore, the sensory hair cells of the saccular epithelium in silver perch are oriented in three-dimensional space, whereas the overwhelming majority of those in other teleosts are positioned in two-dimensional space. This implies that the sensory hair cells on the saccular epithelium of this species are sensitive to a wide range of directional acoustic stimuli. It is possible that the saccule of the silver perch plays a primary role in sound-source localization. The relative functions of the otolithic end organs are thought to be interspecifically variable and overlapping (Platt and Popper, 1981).

Orientation patterns of sensory ciliary bundles of the sacculus have been classified into five categories, including standard, dual, opposing, alternating, and vertical (Popper and Coombs, 1982). The saccular epithelia of non-*Stellifer*-group sciaenids, namely, Atlantic croaker, spotted seatrout, spot, and kingfish, follow the standard pattern (Ramcharitar et al., 2002). The orientation pattern of ciliary bundles on the saccular epithelium of silver perch, as for the bowfin, *Amia calva* (Popper and Northcutt, 1983), a taxonomically distant species, do not easily fit into any of the five groups; they are distinctly three-dimensional structures. Instead, in silver perch, the ciliary bundle orientation on the vertical segment of the saccular macula looks like the vertical pattern found in otophysans and *Gnathonemus* sp., a mormyrid (Popper, 1978, 1981; Platt and Popper, 1981; Popper and Coombs, 1982), whereas the pattern on the other segment of this epithelium has not been reported for any other teleost species because the bundles here are predominantly directed toward the midline.

The vertical pattern of ciliary bundle orientation on the saccular epithelium is correlated with enhanced audition (Platt and Popper, 1981; Popper and Coombs, 1982), and the data for silver perch support the possible role of this arrangement in specialized audition. Along with this, the long axis of the saccular epithelium is parallel to the anterior swim bladder projections that terminate lateral to the otic capsule. It is, therefore, conceivable that particle motion fields set up by resonance of the swim bladder in acoustic pressure fields can potentially stimulate a significant number of hair cells on this sensory epithelium.

If the saccular epithelium of silver perch is flattened, it somewhat resembles the overall "tadpole shape" of other sciaenids (Ramcharitar et al., 2002). It is, therefore, possible that its specializations are derived from an ancestral standard pattern. We propose that the orientation pattern of hair cells on the saccular epithelia of species such as silver perch and bowfin fit into a new category because their epithelia are three dimensional, giving rise to a unique arrangement of ciliary bundles.

Also unique to silver perch is the proximity of the utricle to the saccule as revealed by gross dissections as well as by three-dimensional reconstruction of the otoliths from CT scan analysis (Fig. 5). It is possible that the utricle and saccule here are involved in coupled mechanical dynamics. Further anatomical investigations are needed to determine whether this arrangement confers such dynamics, as well as their possible adaptive value in audition.

The issue of matching between audition and vocalization in fishes is a highly controversial one; the evidence is conflicting for the vast majority of species investigated, e.g., the toadfish and various species of anabantoids (Fine, 1981; Schellart and Popper, 1992; Ladich and Yan, 1998). However, physiological studies have shown that some vocal species may be especially tuned to detect acoustic signatures within their vocalizations (McKibben and Bass, 1999; Kozloski and Crawford, 2000). The study of audition in weakfish, Atlantic croaker, and spot revealed no clear correlation between audition and vocalization (Ramcharitar, 2003) insofar as the energy of their calls is concentrated in frequencies well within 1 kHz, i.e., a frequency detection range common to the vast majority of fishes described in the literature, vocal and nonvocal alike (Fay, 1988). As an exception, there is a correlation between audition and vocalization in silver perch. This species is able to detect sounds up to 4 kHz, and its vocalizations contain significant energy up to several kilohertz (Luczkovich et al., 1999). The adaptive value of the extended frequency range of detection of silver perch may, therefore, be linked to its vocalization and, hence, mate selection.

The anterior segment of the swim bladder of silver perch terminates lateral to the saccules and just beneath the skin, close to a region where networks of the lateral line system exist (Ramcharitar, 2002). This is suggestive of a possible interaction between the swim bladder and the mechanosensory lateral line. In clupeids, the auditory bulla is coupled to the lateral line canal system (Denton and Blaxter, 1976), and it has been shown that vibrations of the bullae cause fluid motion in both the inner ear and the lateral line canals (Blaxter et al., 1981). Webb (1998) showed a functionally similar coupling between the swim bladder and the lateral line in chaetodontid species. To date, however, the biological significance of this rare coupling is entirely unknown.

Recent data on units innervating the superficial neuromasts of plainfin midshipman, *Porichthys notatus*, suggest that they may be responsive to both mechanosensory and acoustic stimulation (Weeg et al., 2002). This is supported by significant overlap between the acoustic and the lateral line ascending pathways in this species (Weeg et al., 2002). It has been suggested that the multimodality of the lateral line system in the plainfin midshipman is related to reproductive behavior of this vocal species (Brantley and Bass, 1994; McKibben and Bass, 1999). Therefore, if a lateral line–swim bladder interaction is shown to exist in silver perch, a vocal species, such an association may also be involved in the receipt of acoustic information during reproductive behavior.

The silver perch thus has unique and diverse anatomical auditory specializations coupled with enhanced hearing ability. Although further work is needed to elucidate the full extent of form and function in the hearing system of this species, the current data strongly support the proposal that members of the family Sciaenidae are excellent models for understanding structure–function relations in the inner ears of fishes and perhaps in vertebrates generally.

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