

Sound Transmission in Archaic and Modern Whales: Anatomical Adaptations for Underwater Hearing

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ABSTRACT

The whale ear, initially designed for hearing in air, became adapted for hearing underwater in less than ten million years of evolution. This study describes the evolution of underwater hearing in cetaceans, focusing on changes in sound transmission mechanisms. Measurements were made on 60 fossils of whole or partial skulls, isolated tympanics, middle ear ossicles, and mandibles from all six archaeocete families. Fossil data were compared with data on two families of modern mysticete whales and nine families of modern odontocete cetaceans, as well as five families of non-cetacean mammals. Results show that the outer ear pinna and external auditory meatus were functionally replaced by the mandible and the mandibular fat pad, which posteriorly contacts the tympanic plate, the lateral wall of the bulla. Changes in the ear include thickening of the tympanic bulla medially, isolation of the tympanoperiotic complex by means of air sinuses, functional replacement of the tympanic membrane by a bony plate, and changes in ossicle shapes and orientation. Pakicetids, the earliest archaeocetes, had a land mammal ear for hearing in air, and used bone conduction underwater, aided by the heavy tympanic bulla. Remingtonocetids and protocetids were the first to display a genuine underwater ear where sound reached the inner ear through the mandibular fat pad, the tympanic plate, and the middle ear ossicles. Basilosaurids and dorudontids showed further aquatic adaptations of the ossicular chain and the acoustic isolation of the ear complex from the skull. The land mammal ear and the generalized modern whale ear are evolutionarily stable configurations, two ends of a process where the cetacean mandible might have been a keystone character. *Anat Rec* 290:716–733, 2007. © 2007 Wiley-Liss, Inc.

Key words: aquatic hearing; sensory systems; evolution; mammal; archaeocete; cetacea; Eocene

Grant sponsor: Ella and Georg Ehrnrooth Foundation; Grant sponsor: National Science Foundation; Grant sponsor: Department of Science and Technology, New Delhi, India.

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Received 2 March 2007; Accepted 6 March 2007

DOI 10.1002/ar.20528

Published online in Wiley InterScience (www.interscience.wiley.com).

Cetaceans descended from terrestrial mammals around 50 million years ago (Thewissen et al., 2001; see also review in Uhen, 2007, this issue). The cetacean sister group is one of the artiodactyls (e.g., Lockett and Hong, 1998; Milinkovitch et al., 1998; Gatesy and O'Leary, 2001; Gingerich et al., 2001; O'Leary, 2001; Thewissen et al., 2001; Geisler and Uhen, 2003), and among the extant artiodactyls, hippopotamids are the most likely sister group of cetaceans (Nikaido et al., 1999; see also Fisher et al., 2007, this issue). The order Cetacea can be characterized by their ears (Berta, 1994). The transition from land to water involved a great number of changes in individual organ systems (see Berta et al., 2006). These different changes happened in less than 10 million years (Thewissen and Williams, 2002). The sound transmission mechanism underwent pervasive anatomical changes that are well understood functionally (Nummela et al., 2004a).

Early whale evolution is well documented in the fossil record (see reviews by Fordyce and de Muizon, 2001; Thewissen and Williams, 2002). The Eocene cetacean families Pakicetidae, Ambulocetidae, Remingtonocetidae, paraphyletic Protocetidae, Basilosauridae and paraphyletic Dorudontidae represent respective higher branches on the phylogenetic tree of Cetacea (O'Leary, 2001; Geisler and Sanders, 2003; Geisler and Uhen, 2003).

Regarding pakicetids, Gingerich and Russell (1981) and Gingerich et al. (1983) provided descriptions of the braincase and the auditory region of *Pakicetus inachus*, and analyzed pakicetid hearing mechanisms in air and water. Their interpretation was that *Pakicetus* was already specialized in aquatic hearing. Oelschläger (1986a,b, 1987, 1990) expanded this work and made further functional inferences. Thewissen and Hussain (1993) described a *Pakicetus incus*, the oldest middle ear ossicle known for cetaceans, and showed that the *Pakicetus* middle ear is land mammal-like and resembles the modern artiodactyl *incus* in relative length of its crura. Luo (1998) and Luo and Gingerich (1999) studied the pakicetid ear structures, and added further details to the functional analyses of the transformation of the cetacean basicranial structures during the evolution of whales. Nummela et al. (2004a, 2006) provided detailed descriptions of the largest collection of cranial material of all three pakicetid genera; the corresponding postcranial material is described by Madar (2007). Regarding ambulocetids, Thewissen et al. (1994, 1996) described this new family while describing the genus *Ambulocetus*. Much of the ear material of *Ambulocetus* is poorly preserved.

Kumar and Sahni (1986) described the ear region of *Remingtonocetus*, within their new family Remingtonocetidae, making suggestions of the hearing mechanism of this species. Nummela et al. (2004a) expanded the study of remingtonocetid ear and hearing, with new cranial material, tympanic bullae, and all three auditory ossicles, and found the remingtonocetids to be the first cetacean family with a whale hearing mechanism for underwater hearing. The sound path from water to the inner ear passed through the lower jaw and mandibular fat pad to the tympanic plate, a lateral wall of the tympanic bone, to the ossicular chain, and the oval window. This mechanism was fully functional, although not yet very sophisticated.

The ear morphology of protocetids has been described for *Georgiacetus* (Hulbert et al., 1998), *Rodhocetus* (Gingerich et al., 1994), *Gaviacetus*, *Takracetus*, and *Dalmanistes* (Gingerich et al., 1995). Luo (1998), and Luo and

Gingerich (1999) described ear morphology of *Gaviacetus* and *Indocetus* and discussed their function and hearing abilities. Nummela et al. (2004a) described the hearing function of protocetids, including *Indocetus* tympanic and malleus. Geisler et al. (2005) described the ear structures and the mandible for *Carolinacetus*. Regarding basilosaurids and dorudontids, Kellogg (1936) described their auditory morphology in detail, but he did not address functional aspects of the fossil whale ear.

The most explicit study of functional character transformations in early whales was by Lancaster (1990), building on earlier work of Fleischer (1978). At the time when Lancaster wrote his study, early cetacean ears with functional interpretations were only known for basilosaurids and dorudontids, and these Eocene whales were included in the hypothetical transformation series he proposed for the whale ear. Lancaster determined that the late Eocene whale ear was a highly derived hearing mechanism capable of sound transmission underwater. Uhen (1998, 2004), Luo (1998), Luo and Gingerich (1999), and Nummela et al. (2004a) added functional interpretations based on the morphology of the lower jaw and the ear region, which indicate that these animals heard well in water, and had already relatively good directional hearing, but did not yet echolocate.

SOUND TRANSMISSION MECHANISMS IN MAMMALS

Although it has long been known that sound transmission through the cetacean ear differs from that of land mammals (e.g., Norris, 1968; Fleischer, 1978), the function of the middle ear was poorly understood until a generalized model for the hearing mechanism of modern odontocetes (toothed whales, including dolphins and porpoises) was designed (Nummela et al., 1999a,b; Hemilä et al., 1999, 2001). This study uses that model to evaluate sound transmission pathways in the Eocene whale ear. It will begin with a review of the acoustic differences between air and water and explain why land mammal ears do not function well in water. Then it will review the three different sound transmission mechanisms found in land mammals and in whales: land mammal sound transmission, modern odontocete sound transmission, and bone conduction. After that, it will describe the morphology and interpret the function of the sound transmission mechanisms found in Eocene archaeocete whales: pakicetids, ambulocetids, remingtonocetids, protocetids, basilosaurids, and dorudontids. The ancestral land mammal ear and the modern odontocete ear represent evolutionarily stable configurations (Wagner and Schwenk, 2000), and together they provide the endpoints of a continuum that is an example of macroevolutionary change in mammalian evolution (Nummela et al., 2004a).

SOUNDWAVE PROPERTIES IN AIR AND WATER

Sound propagation in air and water differs due to differences in physical properties in these two media. Here, we present a brief overview of basic acoustics as background for the functional discussion that follows. Sound velocity c , sound wavelength λ , and sound frequency f are related to each other as: $c = \lambda f$.

Sound velocity in air is approximately 340 m/s, but in water it is nearly five times higher at 1,530 m/s (varying slightly with changes in ambient conditions, e.g., temperature, salinity, pressure). The higher sound velocity in water means that, for a given sound frequency, the wavelength is longer in water than in air. The ratio between the frequency and the wavelength is inverse; when the sound frequency becomes higher, the wavelength becomes shorter, and vice versa. As shorter wavelengths give better spatial resolution, high frequencies are more suitable for detecting small objects than are low frequencies, and are used by many animals in echolocation. However, high frequencies do not carry very far; they attenuate rapidly. Low frequencies have long wavelengths, attenuate more slowly than high frequencies, and thus carry over longer distances. They are more suitable for long-distance communication, whereas high frequencies are better suited for short distances (see e.g., Richardson et al., 1995; Geisler, 1998).

ACOUSTIC IMPEDANCE

Sound propagation in a particular medium can be characterized by its acoustic impedance. When sound moves forward from one medium to another, the ratio between the acoustic impedances of two media dictates how much energy will be reflected at the interface of these media. The larger the ratio, the more energy will be reflected at the interface.

The acoustic impedances of air and water differ greatly. The characteristic acoustic impedance is: $Z = p/v = \rho c$, where p is the sound pressure, v is the particle velocity, ρ is the density of the medium, and c is the sound velocity in that particular medium. The densities of air and water are approximately 1.3 kg/m^3 and $1,000 \text{ kg/m}^3$, respectively, and, as shown above, the sound velocity is approximately five times larger in water than in air. The characteristic acoustic impedance of air is $Z_{\text{air}} = 400 \text{ Pas/m}$, and the characteristic acoustic impedance of water is $Z_{\text{water}} = 1,500 \text{ kPas/m}$. The specific acoustic impedance of the cochlea filled with fluid is approximately one-tenth of the characteristic acoustic impedance of water, and thus $Z_c = 150 \text{ kPas/m}$ (see Hemilä et al., 1995). Hence, the specific impedance of the cochlea is much larger than the characteristic acoustic impedance of air, but again only slightly smaller than the characteristic acoustic impedance of water.

An equal sound pressure gives air molecules a larger particle velocity than water molecules, resulting in lower impedance of air than of water. To overcome an impedance difference between two media, and to decrease the amount of sound reflected at the interface, an impedance matching device is needed. Such a mechanism adjusts the sound pressure and particle velocity, either by increasing the sound pressure, and/or decreasing or increasing the particle velocity between the outer medium and the cochlea (for more details, see the sections on land mammal and modern odontocete hearing mechanisms).

DIRECTIONAL HEARING

The ability to determine the direction from which a sound is arriving is in general species-specific, and also varies according to the sound frequency. For determining the direction of the sound arrival, two different methods

can be used: the interaural time difference, and the interaural intensity difference (see e.g., Masterton et al., 1969; Heffner, 2004). The interaural time difference is the time between the arrival of the sound in left versus right ears. When the sound arrives from front, or from behind, it reaches both ears simultaneously, but by turning the head from one side to the other the animal can tell whether the sound arrives from the front or from the back, by determining the time difference between arrival of the sound at the left and right ears. Whether the interaural time difference can be used for determining the direction of the sound depends on the size of the head, on the direction of the incident sound, and on the frequency of sound. If the interaural distance is large enough in relation to the wavelength of the arriving sound, the animal can determine the direction of the sound on the basis of the interaural time difference (large enough phase difference, however smaller than 2π).

However, when the size of the head is very small, the time between the arrival of the sound in the left and right ears becomes absolutely very short, and no matter how high frequencies are being heard, the central nervous system is not capable of detecting the differences in arrival time. However, the sound intensity is generally larger in the ear where the sound arrives first, and the interaural intensity difference increases with the frequency. Hence, directional hearing based on time difference may be good even at high sound frequencies, but when the sound is high enough, and the animal's head is small, directional hearing is based on intensity differences between the left and right ear, provided that the high-frequency hearing ability is good. This finding partly explains why it is advantageous for a small animal to hear very high frequencies.

Sound travels approximately five times faster in water than in air and, at a given frequency, the wavelength in water is five times longer. This reduces considerably the ability to use the interaural time delay between the two ears for sound localization. However, even with this increase in velocity and wavelength, the high frequencies that odontocetes use have short enough wavelengths in relation to their large head. Hence, the high frequencies together with a large head compensate for more than the fivefold increase in underwater sound velocity. Indeed, odontocetes are known to have very precise directional hearing (see Supin et al., 2001), this being partly a prerequisite for their ability to echolocate.

INTENSITY VS. PRESSURE

In the field of acoustics, the sound pressure is of central importance, because the sound loudness is measured with pressure meters. However, as the perceived loudness is roughly proportional to the logarithm of the rms (root mean square) sound pressure p , the sound loudness, given in decibels dB, is usually expressed using sound pressure level L_p : $L_p = 20 \text{ dB} \log p/p_0$.

The reference pressure value p_0 used for sounds in air is $20 \text{ } \mu\text{Pa}$, and in water $1 \text{ } \mu\text{Pa}$. When comparing hearing sensitivity of different terrestrial or aquatic animals at different frequencies, sound pressure levels are used.

The relevant quantity determining the sensitivity of the cochlea is the incident sound energy. Hence, sound intensity is used when comparing the hearing of a terrestrial versus an aquatic animal. In the case of a plane wave, the

intensity I of the sound wave is: $I = p^2/Z$, where Z is the characteristic acoustic impedance of the medium, Z_{air} or Z_{water} . The ratio $Z_{\text{water}}/Z_{\text{air}}$ is quite large, approximately 3,700. Hence, when comparing plane waves of equal intensities in air and water, the sound pressure in water is larger. Using the conventional reference pressure p_0 values of $20 \mu\text{Pa}$ (for air) and $1 \mu\text{Pa}$ (for water), the waves have equal intensities when the sound pressure level L_p of the aquatic wave is 61.8 dB larger than sound pressure level L_p of the wave in air. Thus, a terrestrial animal and an aquatic animal have equal sensitivities if the threshold L_p value of the aquatic animal is 61.8 dB larger than the threshold L_p of the terrestrial animal.

LAND MAMMAL EAR AND HEARING

The basic function of the land mammal ear in hearing is well understood over a wide range of frequencies (e.g., Henson, 1974; Fleischer, 1978; Fay, 1988; Geisler, 1998; Rosowski and Relkin, 2001). Figure 1A shows a diagram of a generalized land mammal ear. Sound is collected by the outer ear pinna, and travels through the external auditory meatus (EAM) to the tympanic membrane (TyMe), where it sets up vibrations. In the middle ear cavity, the three middle ear ossicles, malleus, incus, and stapes (Mal, Inc, Sta), form an ossicular chain between the tympanic membrane and the oval window (OvW) of the cochlea. The outermost ossicle, the malleus, is attached to the tympanic membrane through its manubrium (Fig. 2A), and the footplate of the stapes sits at the oval window. Malleus and stapes are connected by the incus. The vibrations of the tympanic membrane are transmitted through the middle ear ossicles to the oval window, where the movements of the stapes footplate set the inner ear fluid into motion. The tympanic bone (TyBo) and the periotic bone (Per) have large contacts with each other (PeTy),

and the periotic is in close contact to the squamosum and other skull bones (Sk). As a result, the ear is not acoustically isolated from the skull and sound waves can travel through the skull (see Bone Conduction below). The mandible (Man) is not involved in sound transmission.

When hearing in air, airborne sound is transmitted from lower acoustic impedance of air ($Z_{\text{air}} = 400 \text{ Pas/m}$) to much higher acoustic impedance of the inner ear fluid ($Z_c = 150 \text{ kPas/m}$). In a land mammal ear, reflection of sound is avoided by the insertion of the middle ear which acts as an impedance-matching device between the air and the inner ear.

The impedance matching in a land mammal ear is accomplished by increasing the pressure p and by decreasing the particle velocity v between the tympanic membrane and the oval window ($Z = p/v$, see above). The pressure can be increased by transmitting sound from a larger to a smaller area. This is done with the help of the tympanic membrane A_1 and the oval window area A_2 . The area ratio between these two is A_1/A_2 . The particle velocity can be decreased by having the malleus lever arm l_1 longer than the incus lever arm l_2 . With the lever ratio $l_1/l_2 > 1$ between the malleus and the incus lever arms, the particle velocity between the tympanic membrane and the oval window is decreased. The product of the area ratio and the lever ratio, $A_1/A_2 \times l_1/l_2$, is called the geometric transformer ratio of the mammalian middle ear. With this mechanical transformer, it is possible to minimize reflection of sound energy at the tympanic membrane—in other words, the middle ear input impedance is matched with the input impedance of the cochlea.

In addition to the impedance matching function, the mammalian middle ear also functions as an intensity amplifier. The intensity amplification is accomplished when energy is transferred from a larger to a smaller area, in this case from the tympanic membrane to the oval window. In a land mammal ear, the area ratio between the

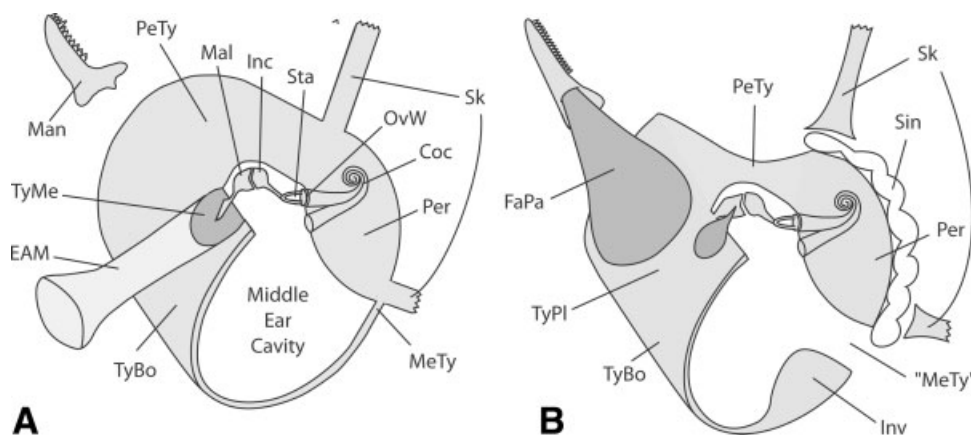


Fig. 1. **A:** Diagram of the land mammal ear. **B:** Diagram of the modern odontocete ear. For technical reasons, the mandibular foramen and the mandibular fat pad are shown on the lateral side of the mandible, although they in reality are situated on the medial side. Abbreviations in Figures 1 and 7: Coc, cochlea; Dom, dome-shaped depression for periotic; EAM, external acoustic meatus; FaPa, mandibular fat pad; Inc, incus; Inv, involucrum; Mal, malleus; Man, mandib-

le; MeTy, medial synostosis between periotic and tympanic bone, in cetaceans this synostosis is absent and is homologous to a gap between these bones ("MeTy"); OvW, oval window; Per, periotic bone; PeTy, joint between periotic and tympanic; Sin, air sinuses; Sk, skull; Sta, stapes; TyBo, tympanic bone; TyMe, tympanic membrane; TyPI, tympanic plate. Reprinted by permission from MacMillan Publishers Ltd: Nature (Nummela et al., 2004a).

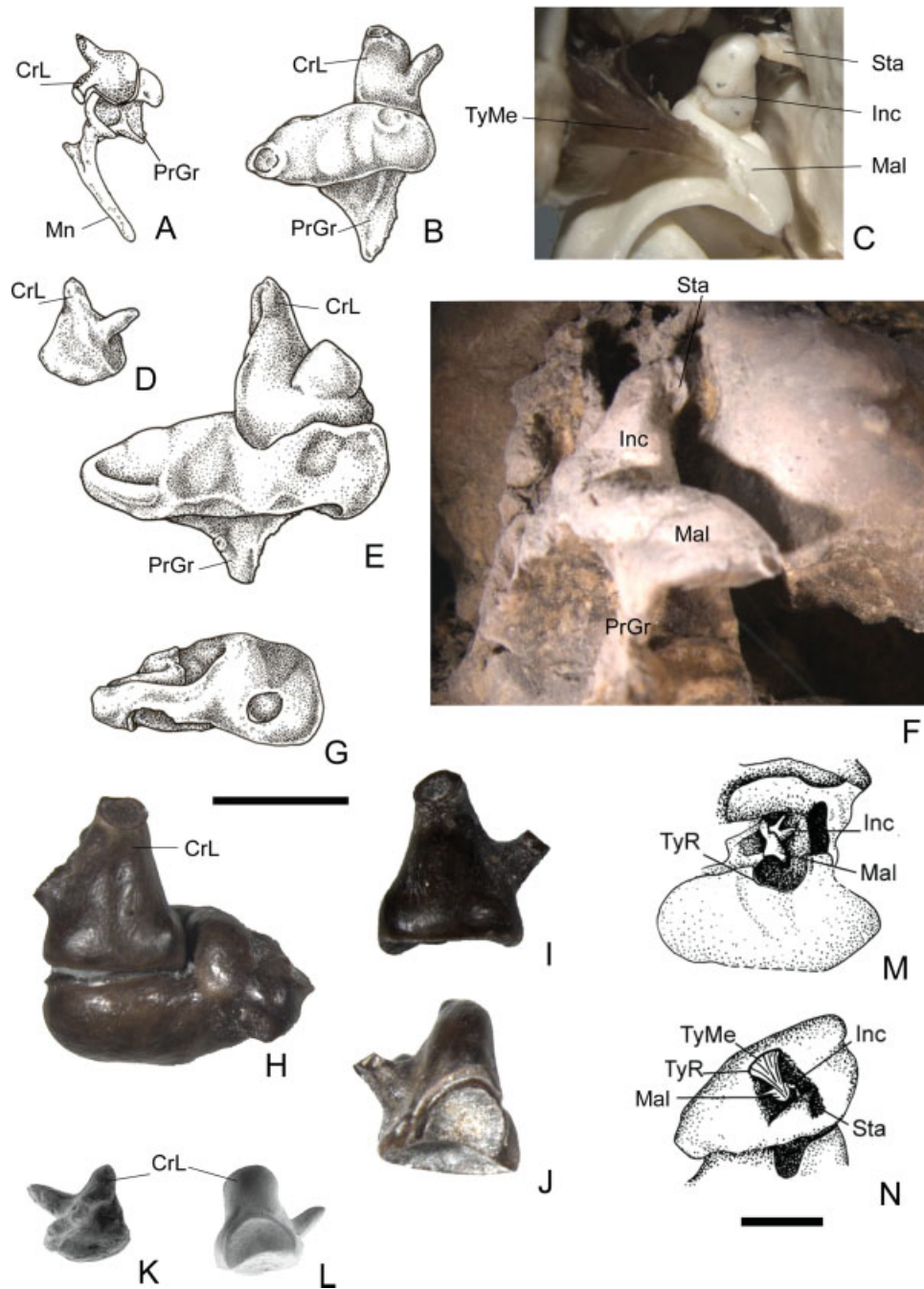


Fig. 2. Middle ear ossicles of fossil and modern mammals. **A:** Left malleus and incus of *Sus scrofa*. **B:** Left malleus and incus of *Lagenorhynchus*. **C:** Left malleus and incus of *Lagenorhynchus* in situ. **D:** Left incus of *Pakicetus* (H-GSP 91035). **E:** Left malleus and incus of *Remingtonocetus* (IITR-SB 2914). **F:** Left malleus and incus of *Remingtonocetus* in situ (IITR-SB 2828). **G:** Right malleus of *Indocetus* (LUVP 11034). **H:** Right malleus and incus of *Basilosaurus*, posterior view (LSUMG V1). **I:** Left incus of *Basilosaurus*, posterior view (LSUMG V1). **J:** Left incus of *Basilosaurus*, anterior view (LSUMG V1). **K:** SEM photograph of the *Pakicetus* incus. **L:**

Right incus of *Lagenorhynchus*. **M:** Left ear of *Remingtonocetus* before the tympanic bulla was removed, showing the ossicles in situ, lateral view. **N:** Ventral view of the same ear, with tympanic membrane reconstructed in the middle ear cavity. Ossicles in A, B, D, E, and G are drawn perpendicular to the plane of the lever arms. Abbreviations: CrL, crus longum; Inc, incus; Mal, malleus; Mn, manubrium; PrGr, processus gracilis; Sta, stapes; TyMe, tympanic membrane; TyR, tympanic ring. Scale bar = 5 mm in A–L; 2 cm in M, N. Adapted by permission from MacMillan Publishers Ltd: Nature (Thewissen and Hussain, 1993), and (Nummela et al., 2004a).

tympanic membrane and the oval window serves two functions: impedance matching and intensity amplification. Moving hardware models originally designed by Tom Reuter and Simo Hemilä for the terrestrial mamma-

lian ear and the modern odontocete ear can be found as video versions on the Web page for whale hearing: http://www.neoucom.edu/DEPTS/ANAT/Thewissen/whale_origins/index.html.

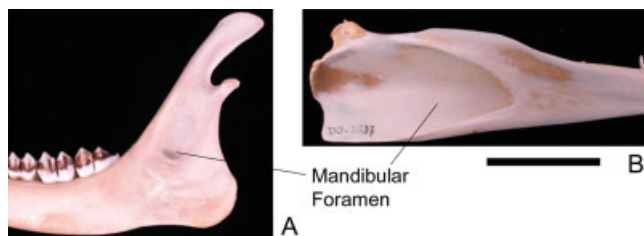


Fig. 3. Medial view of lower jaw showing the mandibular foramen size. **A:** Right mandible of deer, *Odocoileus*. **B:** Left mandible of dolphin, *Lagenorhynchus*. Scale bar = 5 cm.

MODERN ODONTOCETE EAR AND HEARING

The modern odontocete ear is suitable for hearing underwater sound (Reysenbach de Haan, 1957; Fleischer, 1978; see also Oelschläger, 1990; Ketten and Wartzok, 1990; Ketten, 1992, 2000). Figure 1B shows a diagram of the odontocete ear. Modern odontocetes have lost their outer ear pinna, and the external auditory meatus is partially occluded and not functional in hearing. The mandible has a wide mandibular canal, which opens posteriorly as a mandibular foramen on the medial side of the jaw (Fig. 3B). The mandibular canal is filled with a fat pad, an oily substance with a density close to that of sea water (Varanasi and Malins, 1971, 1972). Sound is collected by the lateral mandibular wall (Norris, 1968), and guided through the mandibular fat pad (FaPa) (Fig. 1B) up to the tympanic plate (TyPl), and the lateral wall of the tympanic bone (TyBo), causing it to vibrate. Experimental results support this mandibular sound route at higher frequencies (e.g., Bullock et al., 1968; McCormick et al., 1970; Møhl et al., 1999). However, there is also evidence showing that the head region close to the external auditory meatus is sensitive to lower-frequency sounds (Bullock et al., 1968; Popov and Supin, 1990; Supin et al., 2001).

When sound moves from the mandibular fat pad to the smaller tympanic plate, the intensity is amplified, leading to better sensitivity. In the middle ear cavity, the three middle ear ossicles, malleus, incus, and stapes, form an ossicular chain, as in the land mammal ear. The ossicular chain is situated between the tympanic plate and the oval window of the cochlea, the malleus is connected to the tympanic plate with a bony ridge, the processus gracilis (Fig. 2B). The malleus has lost its manubrium, which in land mammals contacts the tympanic membrane. The tympanic membrane is still left in the odontocete ear, in the form of an elongated tympanic ligament, and it attaches to the malleus with its medial tip (Fig. 2C). The vibrations of the tympanic plate are transmitted through the middle ear ossicles to the oval window (Hemilä et al., 1999; Nummela et al., 1999a), similarly to the land mammal ear. The anatomy of the tympanic bone is different from that of other mammals; the lateral wall is thin, as in mammals in general, but the medial wall is a thick bulky structure, called the involucrum (Inv). The contact between the tympanic bone and the periotic bone (Per) is reduced (PeTy), and thin bony ridges occur on the tympanic side (see computed tomography scans in Nummela et al. 1999a). The whole tympanoperiotic complex is pachyosteosclerotic with a density higher than that found in any other mammal: 2.7 g/cm^3 (Giraud-Sauveur, 1969;

Lees et al., 1983; Nummela et al., 1999b). This bony complex is acoustically isolated from the skull through air sinuses (Sin) between the periotic and the other skull bones (Sk).

In an odontocete ear, waterborne sound is transmitted from higher acoustic impedance of water ($Z_W = 1,500 \text{ kPas/m}$) to lower acoustic impedance of the inner ear fluid ($Z_c = 150 \text{ kPas/m}$). This is a rather small impedance mismatch, leading to an approximately 5 dB sound pressure decrease. However, when the sound vibrations are transmitted from a large tympanic plate area to a smaller oval window, the intensity is increased, which improves the hearing sensitivity, but also the pressure is increased and this leads to a large impedance mismatch. Due to the thin bony ridges between the tympanic and the periotic, and also due to the massive and dense periotic and involucrum, the vibration of the tympanic plate is approximately rotational. In this way, the tympanic plate forms a lever, which reduces pressure and increases the particle velocity. In the ossicular chain, the malleus head together with the incus rotates as a pivot in the epitympanic recess. Here again is a lever with the stapes footplate at the oval window (OvW). This second lever also decreases pressure and increases the particle velocity of the system.

HEARING THROUGH BONE-CONDUCTION MECHANISM

In addition to the land mammal and the modern odontocete hearing mechanisms described above, a third hearing mechanism can be described for mammals: bone-conducted hearing (Lombard and Hetherington, 1993). Bone conduction occurs when sound is transferred from a surrounding medium to the cochlea through vibrations of the soft tissues and bony parts of the head directly, instead of the air passage in the external auditory meatus, or the mandibular fat pad route. A common way to divide between different types of bone conduction is to focus on which anatomical components are mainly involved: outer ear, middle ear, and inner ear; this gives at least three different types of bone conduction (Tonndorf, 1968). Another way is to focus on function; two generally accepted mechanisms for bone-conducted hearing are the compressional and the inertial bone conduction. In the compressional model, a pressure differential develops across the cochlear partition of the inner ear. In the inertial model, relative motion between the ossicular chain and the temporal bone leads to cochlear stimulation much the same way as in air-conducted hearing of land mammals. Both of these models eventually lead to displacement of the basilar membrane, which will create a neural impulse.

Barany (1938) has argued that, in the mammalian middle ear, bone conduction is minimized when the mass center point coincides with the rotational axis of the ossicular chain. This may be of importance when chewing creates a lot of noise, as in terrestrial herbivore mammals. However, bone conduction becomes a useful hearing mechanism in a habitat where the density of the surrounding medium is similar to the density of the body tissues. This means similar impedances, and that sound vibrations can easily penetrate the body tissues, with little attenuation. The density of water is close to the density of soft body tissues, and in water most animals hear through bone conduction, at least to some degree. A nota-

ble exception is the Odontoceti (toothed whales), which have air cushions that isolate the ears effectively from the skull.

The movement of the inner ear fluid in the cochlea caused by bone conduction can even be enhanced by morphological changes in the middle ear ossicles, by moving the mass center point away from the rotation axis, which results in different phases of vibration between the ossicles and the inner ear fluid. This kind of enhancement has evolved in modern mammals, e.g. in phocids and odobenids, in which the incus is inflated (Repenning, 1972; Nummela, 1995), and in some fossorial insectivores and rodents where the malleus is inflated (Cooper, 1928; Stroganov, 1945; Fleischer, 1973; Mason and Narins, 2001; Willi et al., 2006). A disadvantage of bone conduction is that directional hearing is poor when the whole head vibrates, and there is no interaural time or intensity difference available. This is a well-known phenomenon for human divers, too. When an animal's head is under water, bone conduction can only be prevented by isolating the ears from the skull, as is the case in the modern odontocete ear.

In this study, the evolution of underwater hearing in cetaceans is described using fossils. Previous studies of this evolutionary pattern (Fleischer, 1978; Lancaster, 1990; Thewissen and Hussain, 1993) were hampered by the paucity of Eocene whale ear ossicles and the lack of an understanding of the function of the middle ear in modern whales. We here use the functional model of Hemilä et al. (1999, 2001), and Nummela et al. (1999a,b), and expand on the analysis of Nummela et al. (2004a) to build a comprehensive evolutionary model of evolving sound transmission mechanisms during the period when cetaceans became aquatic. Our study does not address the functional changes in the inner ear (but see Ketten, 1992, 2000; Parks et al., 2007, this issue), and leaves open the question of the origin of low-frequency hearing in mysticetes.

MATERIALS AND METHODS

Fossil material was studied for all six archaeocete families. The material consists of whole or partial skulls, isolated tympanics, middle ear ossicles, and mandibles. Mandibles from modern cetaceans were also studied. Comparative data on modern mammalian ears is based on Nummela (1995) and Nummela et al. (1999b, 2004a), as well as new measurements (Table 1). The analysis presented here for the pakicetid hearing mechanisms is based on a large collection of cranial material of pakicetids described by Nummela et al. (2004a, 2006); descriptions for mandibles can be found in Thewissen and Hussain (1998). All the fossil cetacean material studied is listed in Appendix 1.

Parameters Measured

The bicondylar width was measured for our sample of archaeocetes and some modern cetaceans. Data for some additional modern cetaceans were kindly provided by Terry Lancaster and Mark Uhen. Table 1 lists morphometric data for fossil and modern cetaceans. The bicondylar width was used to estimate the body mass for different species. Body size is of great importance as many variables in the skull scale with it. Body size was

estimated following Marino et al. (2004) who found that the bicondylar width correlated strongly with body mass.

Bicondylar width could not be measured on the specimens of the protocetids *Indocetus* and *Babiocetus*. Therefore, bicondylar width was estimated for these taxa by scaling cheektooth/bicondylar width relations in the protocetid *Georgiacetus* (Hulbert et al., 1998).

Measurements were made of the minimum thickness of the lateral mandibular wall along the transect that was located at three quarters of the foraminal length, measured from the posterior end of the foramen (thickness T3 in Nummela et al., 2004b, and in Table 1). Minimum thickness for mandible was determined by running a closed Dyer gauge caliper vertically along this transect and recording the true minimum value of transect. Measurements were also made of the height of the mandibular foramen along this same transect (H3 in Table 1). Additionally, measurements were taken of the bicondylar width and the mandibular foramen height for five non-cetacean mammals: the black bear (*Ursus americanus*), horse (*Equus caballus*), one-humped camel (*Camelus dromedarius*), pronghorn antelope (*Antilocapra americana*), and goat (*Capra* sp.).

The tympanic membrane and the tympanic plate area were determined using a method described by Nummela (1995) and Nummela et al. (1999b), respectively. A Dyer gauge caliper (301-204) was used to measure thickness of the tympanic plate. Tympanic plate thickness is the minimum thickness of this plate as determined by measuring at least five representative areas. The tympanic plate thickness could not be measured for the *Gaviacetus* bulla, as the plate is badly eroded on both sides.

Ossicular mass is a variable of great importance in acoustic studies. Because the specific weight of bone may change during fossilization, a range for the ossicular mass was calculated by determining the volume of the fossil ossicle and multiplying it by the minimum and maximum specific weights known for mammalian bone (land mammals, 2.0 g/cm³, and odontocetes, 2.7 g/cm³; Giraud-Sauveur, 1969; Nummela et al., 1999b; see also de Buffrénil et al., 2004). The minimum and maximum mass values received are both given in Table 1.

RESULTS

The auditory anatomy of the families of archaic cetaceans is described in this section with a focus on comparing the functional components of hearing, not on morphological details. The sound path of the Eocene cetacean families is presented, starting with the mandible and mandibular fat pad (if present), the tympanic and periotic bones, tympanic membrane and/or tympanic plate, and the middle ear ossicles. Expanding on the quantitative analysis on ear ossicle parameters by Nummela et al. (2004a), metric data on these listed elements are summarized in Table 1, and performed as bivariate plots in Figures 4–6. The plots cover the functionally most important aspects of mandibular, tympanic, and middle ear ossicle morphology.

Pakicetids

The mandibular foramen in the lower jaw of pakicetids is small (Fig. 4A), and indicates that no mandibular fat pad was present in the alveolar canal of these earliest

TABLE 1. Morphometric data for cetaceans

	A ₁ (mm ²)	M (mg)	I (mg)	M+I (mg)	TPT (mm)	T3 (mm)	H3 (mm)	BCW (mm)
ARCHAEOCETI								
Pakicetidae								
<i>Pakicetus attockii</i>	350 (41.82)	–	20–27 ¹	–	0.88	1.51	6.88	50.4
<i>Ichthyolestes pinfoldi</i>	–	–	–	–	–	1.45	7.8	42.48
Ambulocetidae								
<i>Ambulocetus natans</i>	–	–	–	–	–	2.4	48.9	76.48
Remingtonocetidae								
<i>Remingtonocetus</i>	922 (83.78)	–	–	200–270 ¹	1.2	1.6	–	82.76
Protocetidae								
<i>Babiacetus</i>	–	–	–	–	–	3.76	54.8	128
<i>Indocetus ramani</i>	1225	144–194 ¹	–	–	1.07	2.8	–	70.12
Basilosauridae								
<i>Basilosaurus cetoides</i>	2295	340–460 ¹	160–216 ¹	500–676 ¹	1.94	–	–	144.8
Dorudontidae								
<i>Zygorhiza</i>	1580	176–373 ¹	120–162 ¹	396–535 ¹	1.55	3.15	101	119
MYSTICETI								
Balaenidae								
<i>Balaena glacialis</i>	5887	1331	516	1847	3.08	–	–	–
<i>Balaena mysticetus</i>	6071	918	407	1317	3.13	–	–	–
Balaenopteridae								
<i>Megaptera novaeangliae</i>	5383	2012	549	2561	2.21	–	–	–
<i>Balaenoptera borealis</i>	5132	2133	850	2983	2.54	–	–	282
<i>Balaenoptera physalus</i>	6508	3077	1230	4307	2.45	–	–	–
<i>Balaenoptera musculus</i>	5979	2303	894	3197	2.23	–	–	435
ODONTOCETI								
Physeteridae								
<i>Physeter catodon</i>	1605	400	324	724	1.23	–	–	–
Kogiidae								
<i>Kogia breviceps</i>	–	–	–	–	–	0.74	47.5	92.75
Ziphiidae								
<i>Ziphius cavirostris</i>	–	–	–	–	–	1.33	66.5	158.1
<i>Hyperoodon ampullatus</i>	1286	351	122	473	1.77	1.35	103	205
<i>Mesoplodon europaeus</i>	–	–	–	–	–	1.51	81	113.2
<i>Mesoplodon bidens</i>	750	156	64	220	1.08	1.15	57.8	103.7
Platanistidae								
<i>Platanista gangetica</i>	930	278	54.2	332.2	1.364	0.3	25	–
Iniidae								
<i>Inia geoffrensis</i>	522	158	34.8	192.8	0.955	2.0	44.5	79.5
Pontoporiidae								
<i>Pontoporia blainvillei</i>	–	–	–	–	–	0.94	24.5	61.5
Monodontidae								
<i>Delphinapterus leucas</i>	525	204	50.5	254.5	0.762	1.84	54.37	142.5
<i>Monodon monoceros</i>	662	260	62.1	322.1	0.866	1.03	66.65	151.9
Delphinidae								
<i>Steno bredanensis</i>	505	143	32.7	175.7	0.744	1.94	38.4	96.74
<i>Tursiops truncatus</i>	539	203	48.8	251.8	0.77	2.03	57.6	115.5
<i>Stenella attenuate</i>	–	–	–	–	–	1.13	32	78.26
<i>Stenella frontalis</i>	318	76.3	19.1	95.4	0.638	2.26	38.64	95.06
<i>Delphinus delphis</i>	348	92.8	21.5	114.3	0.511	1.14	26.72	88.94
<i>Lagenorhynchus albirostris</i>	446	134	38.9	172.9	0.656	1.53	45.28	96.45
<i>Lagenorhynchus acutus</i>	336	92.3	22.1	114.4	0.57	1.24	27.87	83.84
<i>Lagenorhynchus obliquidens</i>	–	–	–	–	–	0.99	38.5	91.78
<i>Lagenorhynchus australis</i>	–	–	–	–	–	1.13	36.95	82.37
<i>Lissodelphis borealis</i>	–	–	–	–	–	0.95	28.29	83.11
<i>Grampus griseus</i>	546	204	52.1	256.1	0.72	2.38	61.5	100.5
<i>Feresa attenuate</i>	–	–	–	–	–	1.15	41.03	81.16
<i>Pseudorca crassidens</i>	741	326	89.6	415.6	1.022	2.8	57.83	151.7
<i>Orcinus orca</i>	1828	757	213	970	1.232	5.48	139	206
<i>Globicephala melas</i>	625	187	55.2	242.2	0.8	1.5	82.95	173.6
Phocoenidae								
<i>Neophocaena phocaenoides</i>	–	–	–	–	–	0.57	23.91	74.58
<i>Phocoena phocoena</i>	333	58.9	14.9	73.8	0.609	0.57	29.5	64.36
<i>Phocoenoides dalli</i>	–	–	–	–	–	0.57	38	81.3

^aDue to possible changes during the fossilization process, minimum and maximum mass values were received for fossil cetacean ossicles; see Materials and Methods.

Data partly collected from: Nummela et al. (1999b, 2004a,b).

A₁ Sound input area; tympanic plate area, for *Pakicetus* and *Remingtonocetus*, also the tympanic membrane area is given in parentheses.

M Malleus mass.

I Incus mass.

M+I Malleus + Incus combined mass.

TPT Tympanic plate thickness.

T3 Minimum thickness of the lateral mandibular wall (see Materials and Methods).

H3 Height of the mandibular foramen (see Materials and Methods).

BCW Bicondylar width.

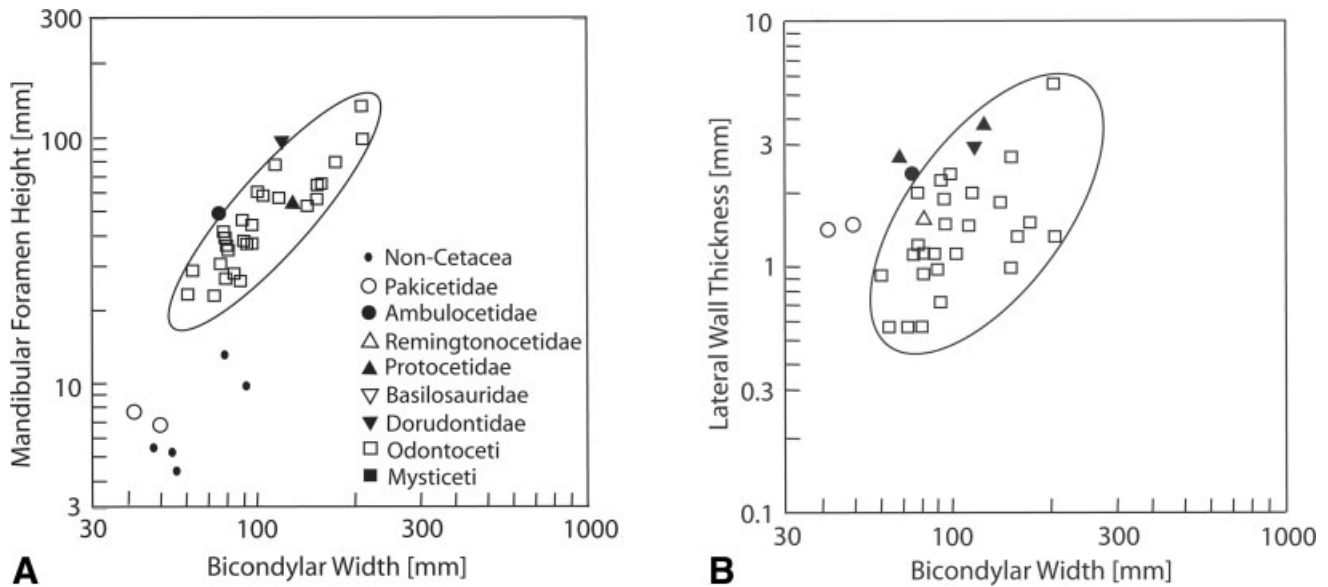


Fig. 4. Variation of the mandibular foramen height and the mandibular lateral wall thickness at the posterior part of the mandible in archaeocetes and modern odontocetes. **A:** Mandibular foramen height vs. bicondylar width. **B:** Lateral wall thickness of the mandible vs. bicondylar width. Odontocete data points clustered within the circle. Symbol chart applies to Figures 4–6.

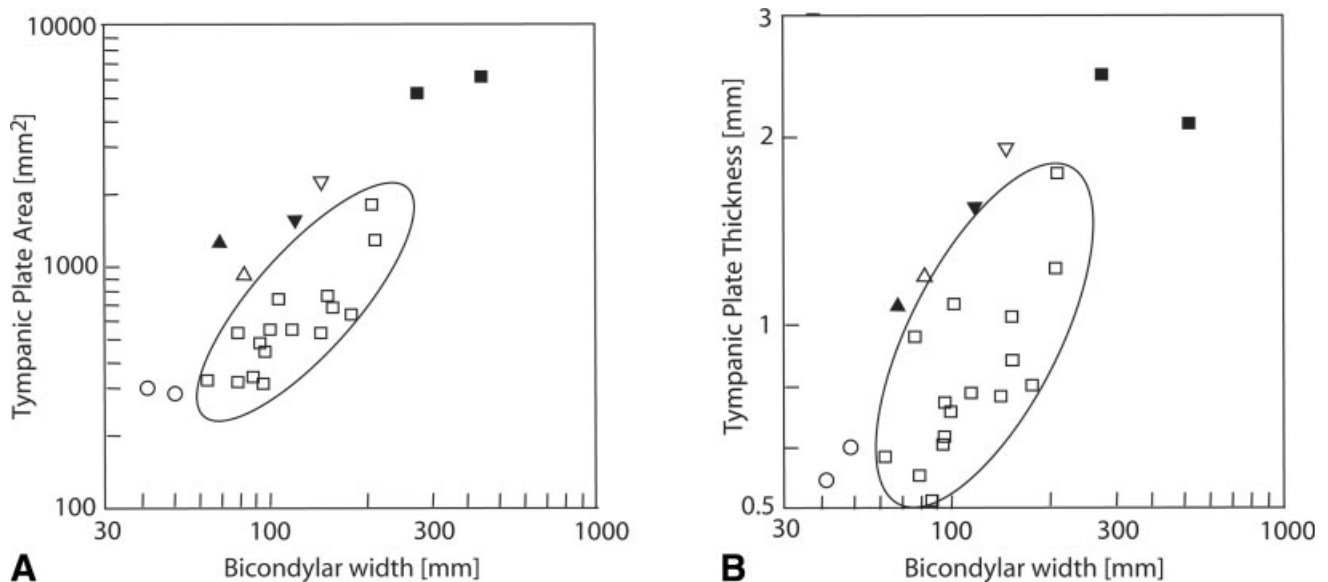


Fig. 5. Variation of the area and thickness of the sound input area of the tympanic bulla, the tympanic plate in archaeocetes and modern odontocetes. **A:** Tympanic plate area vs. bicondylar width. **B:** Tympanic plate thickness vs. bicondylar width. For symbols, see Figure 4A.

whales. The mandibular foramen height of *Pakicetus* and *Ichthyolestes*, rather than following the other cetaceans, falls within a group of noncetaceans including: the pronghorn antelope (*Antilocapra americana*), the goat (*Capra* sp.), the black bear (*Ursus americanus*), the horse (*Equus caballus*), and the one-humped camel (*Camelus dromedarius*), the three first-mentioned being closest to the pakicetids (Fig. 4A). The lateral mandibular wall is also relatively thick (Fig. 4B). In the absence of a mandibular

fat pad as a sound channel to the ear, the lateral mandibular wall could not function as a sound receiver in pakicetid hearing in water.

In all cetaceans, including pakicetids, the tympanic bulla has a thick medial part, the involucrum, and a much thinner lateral wall. The area of the lateral tympanic wall does not as such deviate from the fossil or modern whales (Fig. 5A). The absolute thickness of this lateral wall falls in the same range as that of the smallest

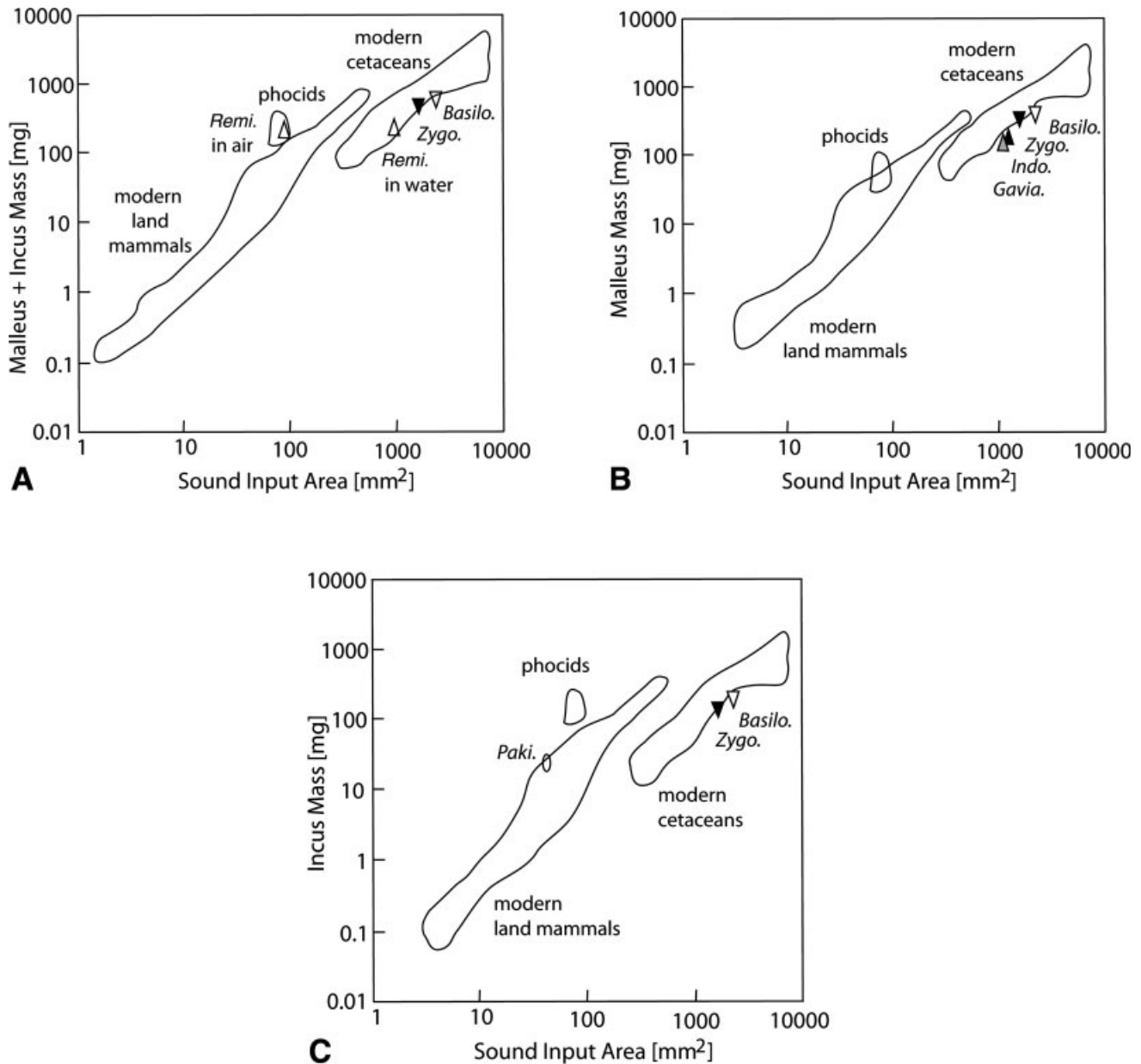


Fig. 6. Variation of middle ear ossicular mass along the sound input area in different mammals. For sound input area, we used the tympanic membrane area for land mammals, seals, pakicetids, and remingtonocetids, and the tympanic plate area for remingtonocetids, protocetids, basilosaurids, and dorudontids. The phocids form their own group by having more massive ossicles than the land mammals, especially the incus. **A:** Combined mass of malleus and incus vs. sound input area. *Remingtonocetus* is shown twice, with its ossicular mass plotted against the tympanic membrane area (hearing in air) and

the tympanic plate area (hearing in water). **B:** Malleus mass vs. sound input area. **C:** Incus mass vs. sound input area. The *Remingtonocetus* could not be included in B and C, because its malleus and incus are not separated from each other. Abbreviated names for the different genera are used, their family level shown by symbols as indicated in Figure 4A. Additionally, following Geisler et al. (2005), *Gaviacetus* is regarded to be a protocetid. Adapted by permission from Macmillan Publishers Ltd: Nature (Nummela et al., 2004a).

odontocetes measured, but when body size is taken into account, the lateral tympanic wall of pakicetids is relatively thick. However, its thickness can be seen to coincide along the thickness line for the Eocene cetaceans (Fig. 5B).

The tympanic bone makes contact with the petiotic bone, which is firmly attached to the skull, with no space

for air sinuses isolating it acoustically from the surrounding skull bones. In the holotype braincase of *Pakicetus inachus* (Gingerich and Russell, 1981), the involucrum of the tympanic bone does contact the basioccipital due to a deformation (Nummela et al., 2006).

The skull morphology reveals that the external auditory meatus was present and patent. On the medial side

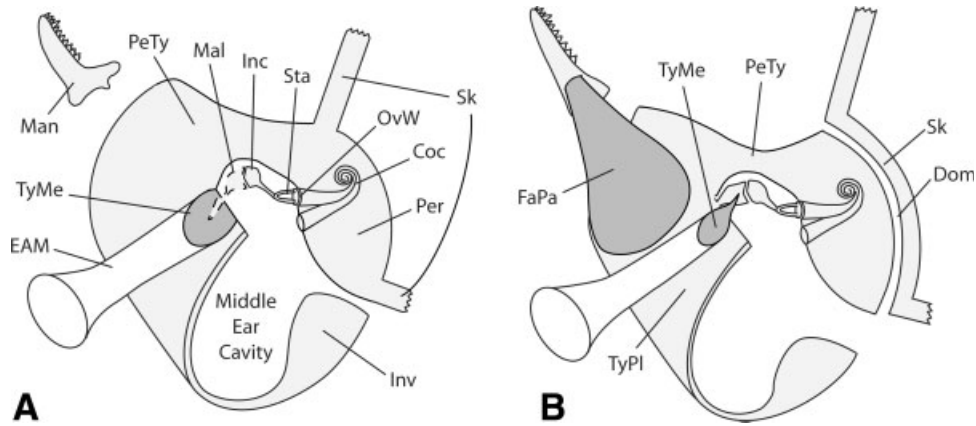


Fig. 7. **A:** Diagram of the pakicetid ear. The malleus is supposed to have been of land mammal type, and its position is shown with a dotted line. **B:** Diagram of the remingtonocetid/protocetid ear. For abbreviations, see Figure 1. Adapted by permission from Macmillan Publishers Ltd: Nature (Nummela et al., 2004a).

of the lateral wall of the tympanic bulla, the shape of the tympanic ring suggests that the tympanic membrane attaching to the ring in life was of the land mammal type. One ossicle, the incus, is preserved for *Pakicetus*. The incus is relatively heavy in relation to the sound input area, but still clusters with modern land mammals (Fig. 6C). The *Pakicetus* incus resembles that of artiodactyls with its crus breve as long as the crus longum (Fig. 2; Doran, 1879; Fleischer, 1973; Thewissen and Hussain, 1993). Generally, the shape of the incus and of the tympanic ring are of land mammal type and indicate that the malleus was of land mammal type too, attaching to the tympanic membrane with its manubrium.

A diagram of the pakicetid ear function is presented in Figure 7A. Results suggest that these earliest whales probably used normal land mammal hearing in air, where sound vibrations reached the tympanic membrane through the external auditory meatus, and were transmitted further by the ossicular chain to the cochlea. Pakicetids most likely used bone conduction for hearing in water, given the close contact between the periotic bone and the skull, and the relatively massive incus. The lack of a mandibular fat pad and the close connection between the periotic and the skull indicate that the lateral tympanic wall was not functionally significant in their hearing mechanism in water, and the modern odontocete hearing mechanism was not present. Directional hearing in water was poorly developed.

Ambulocetids

The *Ambulocetus* head and mandible are massive. The mandibular foramen is large enough to place the *Ambulocetus* among the modern odontocetes (Fig. 4A), and the lateral mandibular wall is relatively thick, at the upper end of modern odontocetes, but well among the other Eocene whales (Fig. 4B). These results suggest that a fat pad was present for sound transmission through the lower jaw to the tympanic bone, although a thick mandibular wall suggests low sensitivity in hearing.

The tympanic bulla has a thick and massive involucrum on its medial side and a thin tympanic plate laterally. In the only known specimen with ears (Thewissen

et al., 1996), the lower jaw and the auditory bulla have a bony contact unlike any other cetacean. Nothing is known of the tympanic membrane and the ear ossicles of *Ambulocetus*.

The contact between the auditory bulla and the lower jaw suggests that hearing of *Ambulocetus* was mainly through bone conduction in water and that it lacked the special modern odontocete hearing mechanism. On land, *Ambulocetus* may also have relied on substrate-borne vibration (Thewissen et al., 1996), by pushing the head against the ground. In water, sound could travel directly from the surrounding water through the tissues to reach the ear. Most likely their hearing was in the low-frequency range; frequencies of around 440 Hz are known to propagate best in soil (Heth et al., 1986).

Remingtonocetids

The lower jaw of *Remingtonocetus* has a large mandibular foramen (Thewissen and Bajpai, 2001a). The lateral mandibular wall is thinner than in pakicetids and ambulocetids, placing remingtonocetids among the modern cetaceans, and indicating synapomorphism between remingtonocetids and later cetaceans in this respect (Fig. 4B).

The external auditory meatus was present. The tympanic bone has a thick medial involucrum and a thin lateral tympanic plate. The tympanic contacts the periotic, which is surrounded by some air sinuses, but still has close contact to the skull. This finding was also noticed by Sahni and Mishra (1972, 1975) in *Remingtonocetus*, which was originally identified by them as a protocetid but later reinterpreted (Kumar and Sahni, 1986; Williams, 1998). The tympanic plate area of *Remingtonocetus* is similar in size to archaeocetes, being larger than in modern odontocetes (Fig. 5A). The tympanic plate thickness shows the same pattern; it is larger than in modern odontocetes, but does not separate remingtonocetids from other archaeocetes (Fig. 5B).

The tympanic ring on the medial side of the tympanic plate and the parts of the plate just lateral to the ring are of land mammal type. This, and the presence of the external auditory meatus, suggest that the sound path for

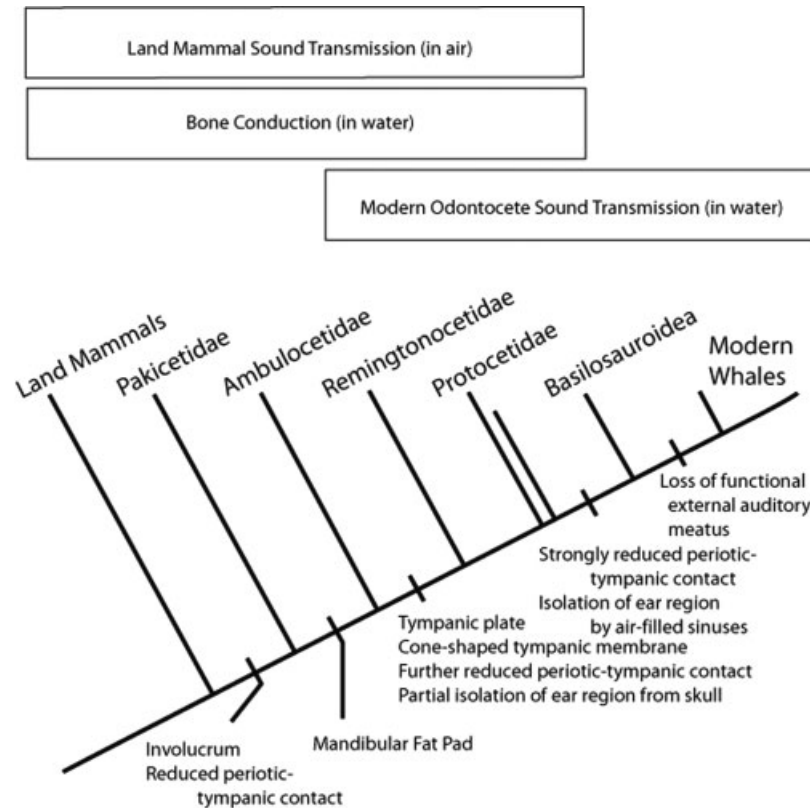


Fig. 8. A generalized phylogeny for the cetaceans discussed in this study, listing the characters relevant to hearing during the evolution of whales. Different sound transmission mechanisms used in air and water by different cetacean taxa are shown in boxes above the

cladogram. The cetacean sister group is here indicated as "land mammals," they used the generalized mammalian sound transmission system. Adapted with permission from Macmillan Publishers Ltd: Nature (Nummela et al., 2004).

air-conducted hearing was still present. The ear ossicles resemble the modern cetacean ossicles morphologically in certain respects. The malleus has a large head and it has lost the manubrium, but has a well-developed processus gracilis, which makes a bony connection to the tympanic plate of the tympanic bulla (Fig. 2). The malleus shape indicates that the tympanic membrane was an elongated ligament that attached laterally to the tympanic ring inside the auditory bulla, and medially to the malleus with its tip. The crus breve and crus longum of the incus are similar in length. Overall remingtonocetid ossicles look like modern odontocete ossicles but minor morphological differences do exist. The ossicular chain is not reoriented. The incus is still relatively large (Fig. 2), unlike in modern odontocetes, where the incus is very small in relation to the malleus.

Possible similarities between the remingtonocetid ear, the land mammal ear, and the odontocete ear were studied using both the tympanic membrane area and the tympanic plate area in the analysis with the middle ear ossicles (Fig. 6). The ossicular mass (malleus + incus) related to the tympanic membrane area in *Remingtonocetus* resembles that of modern phocids (Fig. 6A), who have enlarged middle ear ossicles, and deviate from the general mammalian pattern in this respect (Repenning, 1972; Nummela, 1995). The extra mass of the ossicles most likely helped bone-conducted hearing underwater

by increasing the mass inertia of the ossicles and also by moving the rotational axis and the mass center point apart from each other. These features suggest an increased phase difference between the vibration of the ossicles and the vibration of the inner ear fluid. The ossicular mass related to the tympanic plate area is similar in *Remingtonocetus* and modern odontocetes (Fig. 6A), leading to a functional interpretation that the remingtonocetids had a fully aquatic ear.

A diagram interpreting the function of the remingtonocetid/protocetid hearing mechanisms is presented in Figure 7B. In air, a land mammal hearing mechanism was still present, and sound reached the inner ear through the external auditory meatus, the tympanic membrane, and the ossicular chain. Under water, the modern odontocete sound transmission mechanism was used, with the sound path passing from the lower jaw fat pad to the tympanic plate, and to the middle ear ossicles. The partial connection of the periotic with the skull did not allow fully developed directional hearing in water.

Protocetids

Protocetids have a wide mandibular foramen (Fig. 4A), suggesting the presence of a mandibular fat pad. The lateral wall is thicker than in modern odontocetes, but in accordance with archaeocetes (Fig. 4A). These two features of

the lower jaw indicate that the lower jaw with its fat pad was a functional path in sound transmission underwater.

An external auditory meatus is present in protocetids. The tympanic bone has a thick medial involucrum and a thin lateral tympanic plate. The tympanic makes contacts with the periotic, which is surrounded by air sinuses, but still has close contact to the skull. This arrangement allows for directional hearing to some degree in protocetids. The tympanic plate area of *Indocetus* is larger than in modern odontocetes, as is typical of archaeocetes (Fig. 5A). The tympanic plate is also slightly thicker than in modern odontocetes, but similar to that of other archaeocetes (Fig. 5B). Bajpai and Thewissen (1998) noticed that the shape of the tympanic bulla of remingtonocetids is clearly different from that of protocetids. The most prominent difference between remingtonocetids and protocetids is that, in protocetids, the periotic is more detached from the skull through the surrounding air sinuses. The tympanic and periotic are still in contact with the skull through the basioccipital (Luo and Gingerich, 1999).

A tympanic ring was not identified for the tympanic membrane in the auditory bulla of *Indocetus* or *Gaviacetus*. However, the shape of malleus suggests that the tympanic membrane was an elongated conical ligament, similar to remingtonocetids (Fig. 2). The *Indocetus* malleus is of the modern odontocete type in the lack of a manubrium, and in being attached to the tympanic bulla by the processus gracilis. The shape of the malleus head and the joint facet show that the ossicular chain has reoriented with respect to the position found in remingtonocetids, although it has not yet reached the orientation of the modern odontocete ossicular chain. No incus is available for this group. The mass of the malleus related to the tympanic plate area shows that *Indocetus* and *Gaviacetus* are placed among the modern cetaceans (Fig. 6B).

In air, protocetids used the land mammal hearing mechanism, as suggested by the presence of the external auditory meatus and the connections of the periotic to the skull. Under water, they could hear through the modern odontocete hearing mechanism with their modern whale ear. The bony connection between the ear and the skull, however, indicates that they could not echolocate. Figure 7B describes with a diagram the hearing mechanism of remingtonocetids and protocetids.

Basilosaurids and Dorudontids

The mandibular foramen height in the dorudontid *Zygorhiza* is only slightly larger than the foramen height in modern odontocetes (Fig. 4A). The lateral mandibular wall of *Zygorhiza* is thicker than in modern odontocetes, but is closer to the regression of odontocetes than to that of other archaeocetes (Fig. 4B).

The external auditory meatus is still present in the skull, as in all other Eocene archaeocetes, but its orientation differs from remingtonocetids and protocetids. In basilosaurids and dorudontids, the external auditory meatus is oriented posterolaterally (see also Luo and Gingerich, 1999), whereas in remingtonocetids and protocetids it is oriented laterally. The contact of the tympanoperiotic complex is more derived than in earlier archaeocetes. The tympanic plate area is clearly larger in basilosaurids and dorudontids than in modern odontocetes, following the pattern in other Eocene archaeocetes (Fig. 5A). The tympanic plate thickness in basilosaurids and dorudon-

tids (Fig. 5B) is also similar to that of other Eocene archaeocetes.

The specimens studied did not display a tympanic ring for the possible tympanic membrane inside the auditory bulla, but this finding may be due to the quality of the material. The malleus and incus, especially of *Zygorhiza*, resemble those of modern delphinids and are clearly different from those of remingtonocetids and protocetids (Fig. 2). This finding suggests that the ossicular chain was similar to its position in modern odontocetes, that is, it rotated medioanteriorly, and the tympanic membrane apparently was an elongated conical ligament. Still, the joint facets are relatively larger in basilosaurids and dorudontids than in modern odontocetes, occupying approximately half of the malleolar head length (see also Lancaster, 1990). The incus is relatively larger than in modern odontocetes. However, when plotting the ossicular mass against the sound input area, both *Zygorhiza* and *Basilosaurus* are placed among all other cetaceans, both extinct and modern (Fig. 6).

The footplate area for the stapes of *Basilosaurus* is 9.47 mm² and is an estimate for oval window area (A_2 , see above). With this one example, it seems that the area ratio, the tympanic plate area divided by the oval window area, in *Basilosaurus* is similar to that reported for modern cetaceans by Nummela et al. (1999b).

Basilosaurids and dorudontids hardly used their hearing in air. The strongly reduced contact of the tympanoperiotic complex from the skull suggests that underwater hearing had become the main function of the middle ear. It is unclear what the highest frequencies were that these animals were able to hear; the thickness values of the lateral mandibular wall and of the tympanic plate area do not indicate a very sensitive mechanism.

DISCUSSION

During the Eocene, radical changes took place in the ear anatomy and function of archaeocetes as they adapted to life in water. The modern odontocete ear capable of hearing underwater evolved in less than 10 million years. Our results show that the aquatic ear evolved while hearing on land was still needed, with the origin of remingtonocetids.

The cladogram in Figure 8 shows the characters that are critical for the evolution of an aquatic ear in cetaceans, and the time of appearance of these characters during the Eocene. All the listed characters are necessary for the proper function of the modern odontocete ear. Even so, some of these characters reached their modern state without known intermediates, whereas others underwent gradual changes.

In a modern odontocete ear, relative vibrations between different parts in the tympanoperiotic complex are of crucial importance for the function of this aquatic ear. The degree of the contacts between the tympanic and periotic dictates how these two bones can vibrate in relation to each other. The contacts between the tympanic and periotic bones are reduced already in pakicetids, although to a modest degree. These contacts are further reduced during the Eocene in remingtonocetids and protocetids, in which the first aquatic ear can be found, and in basilosaurids and dorudontids, in which the contacts are still reduced, and close to the odontocete ear.

In water, sound penetrates the body tissues easily due to the small impedance difference. This means that sound can reach the two cochleae almost simultaneously, thus precluding sophisticated directional hearing. In pakicetids, the tympanoperiotic complex is in close contact to the skull. This finding does not affect hearing in air and is a great advantage for hearing through bone conduction in water. However, bone conduction precludes directional sensitivity, as the ears have to be isolated from each other to accomplish this.

One character of the modern aquatic ear already present since the earliest fossil cetaceans is the typically cetacean anatomy of the tympanic bulla, with a thick medial part, the involucrum, and a thin lateral wall. The tympanic bone, with a thick medial involucrum and a thin lateral tympanic plate, is of functional importance for the vibration modes of the tympanic. However, it is plausible that the thick involucrum present in pakicetids and ambulocetids had an important functional role in enhancing the stimulus while hearing through bone conduction under water. The thin lateral wall of the tympanic bone is a plesiomorphic character among mammals in general, but the involucrum is an apomorphy. It is the combination of these that led to the modern cetacean hearing mechanism, although its functional significance in aquatic hearing is evident first in remingtonocetids.

The contacts between the tympanic and periotic, and the acoustic isolation of the whole tympanoperiotic complex from the skull become further reduced in remingtonocetids and protocetids, compared with pakicetids and ambulocetids. This enables the tympanic plate to vibrate more freely, consistent with its active role as a sound-receiving area between the lower jaw and the ossicular chain, and improves the directional hearing of cetaceans. However, both these characters evolve further in basilosaurids and dorudontids, where the contacts between the tympanic and periotic become thinner, and the sinuses around the ear complex increase in size. This change underwent gradual evolution, where full isolation, as seen in modern odontocetes, was not yet reached during the Eocene.

The thin lateral wall of the tympanic, together with the thick medial part, becomes functional in an aquatic ear in remingtonocetids. This takes place simultaneously with the changes in the middle ear structures, the tympanic membrane, and the middle ear ossicles. The tympanic membrane elongates into a conical structure, which then often is called the tympanic ligament. The middle ear ossicles, while still forming the ossicular chain, change in their anatomy so that the ossicular chain becomes slightly rotated, the malleus head enlarges, and its manubrium shrinks to a tiny process to which the tip of the tympanic ligament attaches. The malleus develops a bony contact to the tympanic plate through its anterior process, the processus gracilis, and vibrations from the tympanic plate can now be transferred to the ossicular chain as in an odontocete ear. Additionally, the mandibular foramen in remingtonocetids is large enough to house a mandibular fat pad, a structure that, in an aquatic ear, is a functional analogue to the outer ear of land mammals; the sound vibrations received by the lower jaw can be guided to the tympanic plate of the tympanic bulla. This is even more prominent in *Carolincetus*, the basalmost cetacean of North America; it has an extremely deep

mandibular fossa, apparently housing a large mandibular fat pad (Geisler et al., 2005).

Evolution of the lower jaw and the fat pad as a sound path to the ear was fast and proceeded to its full capacity as soon as this evolutionary invention was available (see review in Uhen, 2007, this issue). However, in some other respects, plesiomorphic characters were retained through the Eocene. For example, all Eocene archaeocetes were diphyodont and heterodont (Uhen, 1998).

The evolution of the tympanic plate of the aquatic ear is more gradual. The plate area is still relatively large and thick compared with the modern odontocetes. A large area collects more energy, but a thinner tympanic plate can vibrate at higher sound frequencies. Also, Ketten (1992) found the *Zygorhiza* to have a mixed high- and low-frequency ancestral type of basilar membrane.

Apparently, high-frequency hearing improved during the Eocene, and this finding can be tied to the evolution of echolocation; decrease of mass enables the system to transmit higher frequencies, which are useful in echolocation. However, based on cranial and cochlear anatomy of archaeocetes as well as fossil odontocetes and mysticetes, Fleischer (1976) suggested that echolocation first evolved among fossil odontocetes, and that no archaeocetes had the ability to echolocate. This is inconsistent with the findings of Uhen (1998) that basilosaurids did not yet show any telescoping of the skull. Directional hearing improved with better isolation of the ears, which is of great importance in developing echolocation ability.

In general, the tympanic plate area and thickness increased in the archaeocetes throughout the Eocene, but then decreased between archaeocetes and the modern odontocetes. While a larger tympanic plate area improves the signal-to-noise ratio, hence improving the hearing sensitivity, a thinner and a smaller tympanic plate has a smaller mass, enabling transmission of higher frequencies. This is relevant for the evolution of echolocation in whales. Characteristic of the anatomical adaptations for underwater hearing is that during their evolutionary history, cetaceans kept the inner ear relatively unchanged (Ketten, 1992, 2000), while they totally reorganized the sound transmission through the outer and middle ear.

ACKNOWLEDGMENTS

We thank the guest editor, Joy Reidenberg, and associate editor, Jeffrey Laitman, for inviting us to contribute to this special issue of *The Anatomical Record*. We also thank Jun Ebersole (McWane Center, Birmingham, AL), James Mead, Charles Potter, and David Bohaska (National Museum of Natural History, Smithsonian Institution), Roger Birkhead and Jennifer Styrsky (Auburn University Museum of Paleontology), Judie Schiebout (Louisiana State University Museum of Geology), and Hans J. Baagøe and Mogens Andersen (Zoological Museum University of Copenhagen) for access to their collections and loan of material under their care. Brooke Garner, Amy Maas, and Lauren Stevens are thanked for their technical assistance with the fossils, and Mary Elizabeth Filon for her artwork. We thank Tom Reuter and Simo Hemilä for designing the hardware models for different mammalian ears, and Rey Notareschi for his expertise in video film techniques. We are indebted to Annalisa Berta, Jason Kosove, Terry Lancaster, Winston Lancaster, and Mark Uhen for helping us collect more

data, and to Simo Hemilä for many critical comments for improving this manuscript. S.N. was funded by the Ella and Georg Ehrnrooth Foundation, J.G.M.T. by the National Science Foundation, and S.B. by the Department of Science and Technology of India.

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APPENDIX 1

Specimens studied for this research are catalogued in the collections of the following institutions:

- AUMP — Auburn University, Museum of Paleontology, Auburn, Alabama (see RMM)
- H-GSP — Howard University, Geological Survey of Pakistan.
- IITR-SB (previously RUSB) — Indian Institute of Technology, Roorkee, India.
- LSUMG — Louisiana State University, Museum of Geoscience, Baton Rouge, Louisiana
- LUVPL — Lucknow University, Vertebrate Palaeontology Laboratory, India.
- NMNH (USNM) — National Museum of Natural History, Smithsonian Institution, Washington, DC.

RMM — McWane Center, Birmingham, Alabama
 VPL — Vertebrate Palaeontology Laboratory, Panjab University, Chandigarh, India.
 ZMUC — Zoological Museum University of Copenhagen, Denmark. The research presented here was based on study of six families of fossil cetaceans, pakicetids, ambulocetids, remingtonocetids, protocetids, dorudontids, and basilosaurids. We here present a list of fossil cetacean specimens studied.

CETACEA

Pakicetidae

Ichthyolestes pinfoldi

H-GSP 18391 (field number 92003), left tympanic with involucrum, lateral plate, sigmoid process, anterior process, and tympanic ring (described by Thewissen and Hussain, 1998; Luo, 1998; Luo and Gingerich, 1999).

H-GSP 30181, right tympanic bulla, rostralateral parts missing.

H-GSP 30223, left tympanic bulla.

H-GSP 30459, right tympanic bulla, dorsolateral parts missing.

H-GSP 91047, right mandible fragment with mandibular foramen (described by Thewissen and Hussain, 1998).

H-GSP 96254, cf. *Ichthyolestes*, juvenile. Braincase and orbital regions with sagittal and nuchal crests, but lacking basicranium and squamosal.

H-GSP 96283, left tympanic with involucrum and fragment of tympanic plate.

H-GSP 96407, left temporal region with periotic and cochlea preserved in part as endocast, external auditory meatus and epitympanic recess.

H-GSP 96431, fragment of left basicranium, which includes periotic with promontorium and oval and round windows, and tympanic with involucrum and lateral plate.

H-GSP 96623, partial skull with right ear region. Periotic with promontorium and epitympanic recess. Squamosal with external auditory meatus, anterior facet, and mastoid process (briefly described by Nummela et al., 2006).

H-GSP 98134, skull including braincase and both ear regions with promontorium, oval and round windows, external auditory meatus (briefly described by Thewissen et al., 2001).

Nalacetus ratimitus

H-GSP 30325, right tympanic bulla, medial part (involucrum).

H-GSP 96384, left tympanic, part of lateral wall missing.

H-GSP 96385, left tympanic, part of ventrolateral wall missing, but with internal cast.

H-GSP 96386, braincase with left squamosal and ear region, promontorium, epitympanic recess, and external auditory meatus.

H-GSP 98140, internal cast of tympanic bulla.

Pakicetus attocki

H-GSP 1649, mandible (holotype, see West, 1980).

H-GSP 30386, left tympanic with involucrum and lateral plate.

H-GSP 91034, right mandible with mandibular foramen (described by Thewissen and Hussain, 1993, 1998).

H-GSP 91035, left tympanic with involucrum, lateral plate, and tympanic ring; left incus.

H-GSP 92100, left tympanic with involucrum, lateral plate, and anterior process.

H-GSP 96192, left tympanic with involucrum and parts of lateral plate.

H-GSP 96231, skull including braincase and right ear region with promontorium, oval and round windows, external auditory meatus (briefly described by Thewissen et al., 2001).

H-GSP 96344, left tympanic with involucrum and lateral plate (published with transposed numbers as H-GSP 96334 by Luo, 1998; Luo and Gingerich, 1999).

Pakicetidae indet.

H-GSP 30119, left tympanic with involucrum and lateral plate.

H-GSP 30120, left tympanic with involucrum and parts of the lateral plate.

H-GSP 30142, right tympanic with involucrum and lateral plate.

H-GSP 30150, right tympanic with involucrum and parts of the lateral plate.

H-GSP 30198, pakicetid n. sp. Right tympanic bulla, caudal and caudomedial parts missing.

H-GSP 30379, right tympanic with lateral plate.

H-GSP 30387, right tympanic with involucrum and lateral plate.

H-GSP 30389, left tympanic with involucrum and lateral plate.

H-GSP 30398, unassociated parts of tympanic bulla.

H-GSP 30424, right tympanic with involucrum and lateral plate.

H-GSP 96506, left tympanic with involucrum and lateral plate.

H-GSP 96541, right tympanic with involucrum, lateral plate missing, but with internal cast.

H-GSP 98138, right tympanic with involucrum and lateral plate.

H-GSP 98141, right tympanic with involucrum and lateral plate.

H-GSP 98143, internal cast of left tympanic bulla.

H-GSP 98199, right tympanic with involucrum and parts of the lateral plate.

H-GSP 98220, right tympanic bulla, rostral and dorsal parts missing.

Ambulocetidae

Ambulocetus natans

H-GSP 18507, skull, partly damaged and mediolaterally compressed, right tympanic with fragments of dentary and basioccipital, left tympanic with parts of mandibular condyle, left posterior mandible (this specimen described in detail by Thewissen et al., 1996).

Remingtonocetidae

Remingtonocetus sp.

RUSB 2521, left and right mandible with mandibular foramen (dental morphology published by Thewissen and Bajpai, 2001b).

RUSB 2592, left mandible with mandibular foramen (dental morphology published by Thewissen and Bajpai, 2001b).

RUSB 2781, right ear region with parts of squamosal, periotic, parietal, supra- and exoccipital, including the external

auditory meatus. The promontorium with oval and round windows, epitympanic recess, and internal auditory.

RUSB 2828, braincase, left tympanic with involucrum, tympanic plate, and sigmoid process; promontorium; the ossicles in situ: malleus with gonial process, incus with short and long processes, and stapes.

RUSB 2914, left tympanic with involucrum, tympanic plate, sigmoid process, posterior process and tympanic ring; malleus with gonial process; incus with long and short processes found in anatomical contact.

Andrewsiphius sp.

RUSB 2786, right ear region with parts of squamosal, periotic, parietal, supra- and exoccipital; promontorium with oval and round windows, epitympanic recess, and external auditory meatus.

Protocetidae

Indocetus ramani

LUVF 11034, right tympanic with involucrum, lateral plate, and sigmoid process; right periotic with promontorium, oval and round windows, facial canal, internal auditory meatus; right malleus with joint facet for the incus, but gonial process missing (described by Sahni and Mishra, 1975; Kumar and Sahni, 1986; Williams, 1998; Luo, 1998; Luo and Gingerich, 1999; Theissen and Bajpai, 2001a; Spoor et al., 2002).

Babiacetus

RUSB 2512, right mandible, posterior part with M₁-M₂, and mandibular foramen.

Gaviacetus

RUSB 2870, left tympanic with involucrum and lateral plate, left malleus with partly broken joint facet for the incus, gonial process missing.

Basilosauroidae

Dorudontidae

Zygorhiza kochii

LSUMG V160A, left tympanic with involucrum and tympanic plate, left incus with short and long processes (described by Lancaster, 1990).

RMM 2739, left tympanic bulla and left malleus (described by Lancaster, 1990 as AUMP 2368).

USNM 4680, right tympanic bulla (described by Kellogg, 1936).

USNM 4748, right tympanic bulla (described by Kellogg, 1936).

USNM 11962, skull (holotype), right and left tympanics, right and left mandibles (described by Kellogg, 1936).

Basilosauridae

Basilosaurus cetoides

LSUMG V1, right malleus with gonial process, right and left incus with long and short processes, right stapes (described by Lancaster, 1990).

RMM 2740, left tympanic bulla.

USNM 4677, left tympanic bulla, dorsal and lateral parts missing (described by Kellogg, 1936:24).

USNM 6125, left tympanic bulla (described by Kellogg, 1936:24).