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A single-ossicle ear: Acoustic response and mechanical properties measured in duck

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ABSTRACT

To date, the single-ossicle avian middle ear (ME) is poorly understood, despite its striking resemblance to the design of many currently used ossicular replacement prostheses. This study aims to improve comprehension of this system. The acoustic response and the mechanical properties of the mallard middle ear were studied by means of optical interferometry experiments and finite element (FE) simulations. A finite element model was constructed based on μCT data and validated using the experimental results. Stroboscopic holography was used to measure the full-field displacement of the tympanic membrane (TM) under acoustic stimulation, and the transfer function was obtained with laser Doppler vibrometry. A sensitivity analysis concluded that the most influential parameters for ME mechanics are the elasticity of the TM, the extracolumella (the cartilaginous part of the columella) and the annular ligament of the columellar footplate. Estimates for the Young's modulus of the TM were obtained by iteratively updating the FE model to match experimental data. A considerable inter-individual variability was found for the TM's elasticity. Comparison of the experimental results and the optimized FE model shows that, similar to the human middle ear, damping needs to be present in the TM to describe the specific spatial and frequency dependent vibrations of the TM. In summary, our results indicate which mechanical parameters are essential to the good functioning of the avian ME and provide a first estimation of their values.

1. Introduction

The avian middle ear (ME) uses the tympanic membrane (TM), a cartilaginous unit (the extracolumella) (EXC) and a single ossicle (the columella) to bridge the acoustic impedance difference between air and the fluids of the inner ear (IE). Although the hearing frequency range in birds is generally smaller than in mammals, with an upper frequency limit around roughly 10 kHz (Dooling et al., 2002), hearing thresholds in both classes are mostly comparable. Just as in mammals, impedance matching in avian species is obtained by three mechanisms: (1) a hydroacoustic transformation represented by the TM-to-footplate area ratio, (2) a mechanical lever action based on rotations around a fulcrum (Saunders et al., 2002), and (3) a curved membrane effect. The mechanical lever is supposed to go along with a tilting motion of the columella and the footplate (FP), given the acute angle between the ossicle and the TM plane (Gaudin, 1968), although detailed measurements are missing. A piston-like motion has been reported in one owl species, which was attributed to an additional flexing motion of the EXC (Norberg, 1978).

It is not well understood how the avian ME deals with quasi-static pressure changes. It has been suggested that the intracolumellar joint plays a role by performing a buckling motion, which is identified as a synchondrosis that functions as a ball joint (Mills and Zhang, 2006; Arechvo et al., 2013). To understand this quasi-static and acoustic behavior, a thorough knowledge is needed of the mechanical parameters that describe the columellar bird ear.

Abbreviations: (μ)CT, (micro) computed tomography; AL, annular ligament; C, columella; ES, extrastapedial process; EST, extrastapedial tip; EXC, extracolumella; FE, finite element; FP, footplate; IE, inner ear; IS, infrastapedial process; MDT, middle drum-tubal ligament; ME, middle ear; S1,2,3, sample 1, 2 or 3; SPL, sound pressure level; SS, suprastapedial process; ST, soft tissue structures; TM, tympanic membrane

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Understanding the functioning of the single-ossicle avian ear may eventually contribute to the improvement of current single-ossicle prosthesis.

Studies of the mechanical properties of the avian ME are scarce, and only describe (quasi-)static characteristics (e.g. Thomassen et al., 2007). Moreover, a priori knowledge of material parameters is lacking entirely. In this study, the acoustic response and mechanical properties were investigated in mallard duck (*Anas platyrhynchos*) by means of optical interferometry and finite element (FE) modeling. Sound-induced motions of the TM were measured with stroboscopic holography to obtain its full-field displacement, and the sound-induced velocities of the columellar footplate and the conical tip of the TM were measured using a laser Doppler vibrometer. A 3D FE model of a duck's ME is constructed, based on the geometry obtained from μCT scans. Using this model, the mechanics of the ME are simulated under acoustic stimulation of the TM. Since initially defined material parameters are uncertain, a sensitivity analysis is performed to quantify their relative influence on the model output. Afterwards, the most influential parameter is determined in inverse analysis for different specimens, which allows us to study the acoustic behavior and viscoelastic properties of the TM at multiple frequencies.

2. Methods

2.1. Experiments

2.1.1. Sample preparation

Measurements were performed on three dissected left ears of defrosted mallard duck heads (S1, S2 and S3). S1 and S2 were male and S3 female. Under the operation microscope, no signs of pathology were detected. During preparation, a part of the left side of the skull containing the ME was dissected from the head, which opened the bilaterally connected middle-ear cavities. The quadrate, which is a part of the beak suspension connected to the ME, was partially removed and the major part of the ear canal was drilled away to expose the TM. After a first set of measurements, the IE load on the ME was removed by drilling away its medial wall and draining the IE fluid. The samples were kept moist by use of a vaporizer (Bionaire) and by putting them in hydrated paper between the preparation and the measurements. In between measurements, samples were stored in refrigerated saline solution.

2.1.2. Stroboscopic holography

Digital stroboscopic holography enables the quantitative measurement of full-field displacement of a vibrating object as a function of time. This is realized by synchronizing very short laser pulses (8 ns) to the vibration phase so that the object's motion is ‘frozen in time’. The full-field displacement at the chosen phase is then calculated by comparing the displaced hologram to a reference hologram of the object in rest. By cycling the laser pulses stepwise through the vibration period at evenly-spaced phase instants, the entire time-resolved transverse motion of the surface is obtained. After Fourier analysis of the time-dependent displacement waveforms, the displacement magnitude and phase maps can be obtained. The exact phase difference between the incident sound waves and the vibrating surface is monitored with an oscilloscope. For a more detailed description of this technique, see Cheng et al. (2010, 2013), Khaleghi et al. (2013) and De Greef et al. (2014a). Sound pressures of 11 frequencies ranging from 0.05 to 12.8 kHz, two per octave, were applied to the lateral side of the TM with pressure amplitudes between 90 and 110 dB SPL. The actual sound pressure at the TM was recorded with a probe microphone. During the measurement, the samples were placed inside a fixture with the TM plane positioned perpendicular to the illumination beam of the laser. On S3, the motion was measured with both intact and removed IE to measure the effect of the IE impedance on the TM response. To enhance reflectivity of the TM, the membrane was painted with a thin layer of either of two different coatings: a suspension of 5% TiO2 in deionized water for S1 and white make-up liquid (Kryolan Aquacolor Soft Cream - White Wet Make-up, Product Code 01129/00; Kryolan, Berlin, Germany) for S2 and S3. Tests showed that the latter gives the best combination of reflectivity, ease of application and delay of dehydration.

2.1.3. Laser Doppler vibrometry

The sound-induced motions of the ossicle were measured on S2 and S3 using a laser Doppler vibrometer (OFV-534, Polytec, Waldbronn, Germany) that is mounted on a surgical microscope (OPMI Sensera/S7, Carl Zeiss, Jena, Germany). Sound-induced velocities were divided by the sound pressure measured in front of the TM to define the middle-ear transfer functions. To enhance reflectivity, a little piece of reflective tape is placed onto the point of measurement, small enough to minimize inertial effects. During measurement, the laser beam was pointed perpendicular to the object's surface. Pure tone sinusoidal pressures, 16 per octave, with amplitudes of 90 dB were presented to the lateral TM surface. A probe microphone was placed in front of the TM to measure the actual pressure. Experiment control and signal processing was done in Matlab. Since it was not feasible to have optical access to the footplate from the lateral side, the following approach was applied: first, the velocity response was measured at the conical tip of the TM from the lateral side, with IE intact. Then, the IE was opened and drained and the transfer function was measured again at the TM to examine the effect of the IE impedance. Finally, the response was measured at the FP in the oval window from the medial side.

2.2. Finite element modeling

2.2.1. Morphology

The geometry for our FE model is based on μCT images of a dissected left ear of a mallard duck, different from the ones used in the experiments. The μCT scan was executed at the University of Ghent Computer Tomography (UGCT) facility (Masschaele et al., 2007). To enhance soft tissue contrast, the ME sample was stained during two days before scanning using a daily refreshed 2.5% PTA solution in deionized water, which limits tissue shrinkage most (Buytaert et al., 2014). The resulting dataset is built up of 2000 × 2000 × 1640 cubic voxels with a voxel size of 7.5 μm. Image segmentation of the CT data was carried out in Amira® 5.3 (FEI Visualization Sciences Group, Hillsboro, Oregon, USA). An automatic seed fill algorithm was applied together with an interpolation method to obtain the segmentation, although manual intervention was required to detect boundaries of soft tissue structures. After segmentation the different geometric components were converted separately into triangulated surface objects (STL), which were recombined in FE software (COMSOL® Multiphysics 5.0, Burlington, Massachusetts, USA). The final triangulated surface contains the ME structures shown in Fig. 1, but the ear canal, the ME cavity wall and the IE were not considered, in order to not overcomplicate the model and to obtain well-defined boundary conditions. The geometry includes the following objects: a slightly conical TM with the apex pointing outwards into the ear canal, the columella bounded by an annular ligament, the EXC considered as a single unit comprising three arms (the infra-, extra- and suprastapedial processes), and Platner's ligament made of collagen fibers which extends across the ME cavity onto the otic process of the quadrate (Starck, 1995). The extrastapedial process ends in the apex of the conical TM which is referred to as the extrastapedial tip (EST). The

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